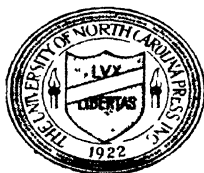


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No. 1

SOUTHERN APPALACHIAN GRASS BALDS

By B. W. WELLS

PLATES 1-5 AND 2 TEXT FIGS.

INTRODUCTION

Scattered at infrequent intervals on the high ranges of the Southern Appalachian mountains are local openings in the forest, the central areas of which are dominated either by a shrub community or an herbaceous grass or sedge community. All such openings which, unlike clearings in the lower mountains, are relatively stabilized or permanent, have, from the earliest historical times, been called "balds." Those which are dominated throughout by a shrub complex, with grass or sedge confined to cattle or man-made trails, are shrub balds (if compact, locally known as "slicks") and are highly variable in size, composition, and site. In contrast, the grass and sedge balds exhibit a central area of varying size with the herbaceous perennials almost completely dominant and shrubs if present constituting a border community transitional to the environing forest. These grass balds are of restricted size (1-100 acres), and exhibit little diversity in composition and site. Roan Bald of a thousand acres is an exception having been greatly enlarged in the modern period. Both types of openings are called "balds" by the local mountain people and the government topographic maps. It is very important therefore to point out that in this paper we are dealing only with the grass bald (including for purposes of simplification the few closely related sedge types) which may always be recognized by the relatively high local dominance of the grasses or sedges on areas generally central, yet immediately surrounded by shrubs or in some directly by forest.

HISTORICAL

The grass balds have been an ecological puzzle to naturalists and apparently to the Indians as well. The Cherokees held to supernatural explanations. The white observers up to the present have insisted on "natural" causes in the sense of local impact of extreme climatic conditions and uncontrolled fire.

CHEROKEE LEGENDS

Because of their human interest and as evidence of the antiquity of the balds, the Cherokee legends deserve restatement here. Fink's (8) account of the most elaborate myth from the original report by Mooney (15) is given first:

"It seems that in the ancient days the Indian villages were subject to the incursions of a mythical monster, an ulagu, resembling a gigantic hornet, that would swoop down, snatch up a child in its claws and vanish so swiftly that pursuit was impossible

"Every possible method of defense and offense was tried, with no avail. Meantime the raids continued, and the villages were fast being depopulated. At last sentinels were posted on the mountain tops and by this means the ulagu was finally traced to its lair, an inaccessible cavern high on a sheer, precipitous mountain side.

"While they had found its retreat, the Indians were little better off than before. In their extremity, they gathered together in a great council and implored Divine assistance. The Great Spirit heard their pleas, and sent to their aid the lightning, that at one tremendous stroke split off the whole side of the mountain. When the smoke and dust cleared away, there lay the ulagu, dazed but still alive. Quickly the Indians fell upon it with spear and ax, ridding themselves forever of the dread scourge. So pleased was the Great Spirit with their initiative in uncovering its hiding place, their piety in appealing for Divine aid in their extremity, and their bravery in the final combat, that it was His decree that in the future the tops of the highest mountains be bare of timber, to better serve as stations for sentinels should such another visitation ever occur."

In the original report the ulagu (Tsul 'kalu or Jutaculla) was definitely located on and in the region of Tennessee Bald which is one of the locally known "Jutaculla Fields" comprising the neighboring two Rough Butt balds together with Gage and Charley Bald.

On Joanna Bald on the Graham-Cherokee County line (not given on topographic maps) was the "lizard place" where a great lizard with glistening throat could be seen sunning himself.

Still another story relates that the balds were forbidden territory and a party of young braves boldly and in merriment entered the open ground of one, whereupon the devil in the form of a huge snake assaulted and swallowed fifty of them.

In the Mooney (15) report mention is made of Gregory Bald which the Cherokees called "the rabbit place" where the king of the rabbits lived—"a rabbit as big as a deer." Near Robbinsville is located a mountain having two small balds which were formed by "a mysterious being having the form of a giant with head blazing like the sun once alighted here and stood for some time. Later they found the herbage burned from the ground."

Another legend states that the Nunnehi (people who live anywhere), a race of spirit folk who inhabited the high mountains, "had a great many town houses on the balds ("udawagunta"—Cherokee name for bald mountains).

RECENT THEORIES

William Bartram who traveled in the Southern Appalachians in 1776, and the Michauxs, father and son, the first white naturalists to traverse the Southern Appalachians make no mention of the balds. They seem to have kept to the main lowland trails.

Gray (9) visited Roan Mt. Bald in 1841 but was not aware of the problem, since the extensive grazing probably led him to believe the entire bald was a mountaineer clearing. His plant list constitutes a valuable record.

Edson (7) is the first writer to attempt a scientific explanation. From observations on Roan she developed a theory of ice storm injury. The ice formation at certain places is so great, with the consequent repression of the trees, that grass comes to take the place of the injured woody plants.

Harshberger (10) naïvely adopts the Edson theory *in toto*.

Davis (6) describes the "grassy balds" of the Black Mt. range but offers no theory. There is "no legend or evidence of Indian occupancy."

Fink (8) under the title, "A Forest Enigma," holds that no theory yet advanced is adequate. He mentions the possibility of Indian origin only to point out what appear to him as unanswerable arguments against this idea. "Nor could they have been cleared by the Indians in pre-historic days for it would have been too gigantic a task, far beyond the possibility of their rude stone axes. Indian energy, or rather lack of it, would not have gone to such incredible labor without some vitally

compelling cause. And even had they done so in the centuries which must have elapsed, the clearings would have been completely reforested."

Camp (3) from observations on one bald; viz., Gregory Bald, believes the balds due to "local exposure to hot and dry southwesterly winds in dry seasons—a local climate theory comparable to the ice storm idea mentioned earlier.

Cain (1) goes no further than to assert that the grass balds must be "natural" phenomena but does not attempt to outline what the differential natural complex is which has produced them.

Wells (17) discusses the problem but offers no theory. In a later preliminary note (18) he calls attention for the first time to the artificial nature of the grass balds since a high percentage of them occupy gentle south slopes near springs, which data together with observations in successional relations, point toward an Indian theory of origin. The present paper is an elaboration of this position.

Wells (19) gives an account of Andrews Bald, using this bald as a basis for a very brief emphasis on the prehistoric human origin of that bald.

DESCRIPTION OF GRASS BALDS

Up to the present the isolated grass balds have not been seriously affected by modern civilization. The Federal high mountain roadway program will soon bring so many people to them that profound changes are certain to follow. In places such as Tennessee Bald the road will traverse the bald itself which will mean its destruction. It is thus highly desirable to record the salient facts concerning these ecologically interesting areas before their vegetation is destroyed in whole or in part.

Because of the isolation of the areas, frequent visits were impossible, and time on the balds was highly limited. No claim is made for completeness of the plant list, especially of the minor species. The few dominants, which are the significant plants in area control, have been very completely recorded.

In the bald descriptions to follow, the plants in the grass bald areas proper are not only listed but ranked as to dominance on a 1-5 scale as follows: 1, dominant; 2, subdominant; 3, frequent or common but not subdominant; 4, uncommon but not rare; 5, rare or a few specimens only present. Plants attaining ranks 1-3 are few in number and are listed in the description with rank 4 and 5 plants listed by number referring to the plant list given below. So far as possible Britton and

Brown's Flora of the Northern States and Canada has been followed in the matter of nomenclature. The location, altitude, month and year the bald was observed and approximate size are given for each bald, followed by the concise description.

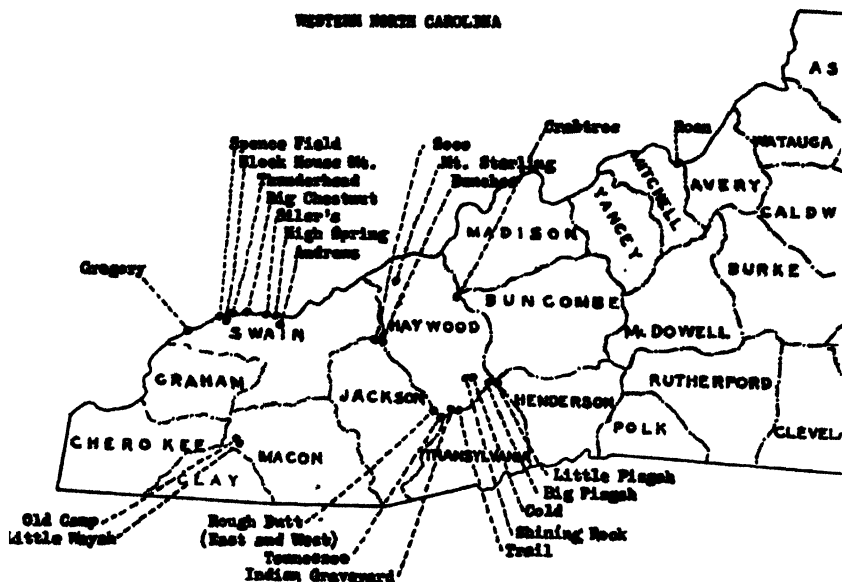


FIG. 1. LOCATION OF GRASS AND SEDGE BALDS DESCRIBED

PRELIMINARY GRASS BALD PLANT LIST

- | | |
|---------------------------------|---------------------------------------|
| 1. <i>Achillea millefolium</i> | 17. <i>Carex flexuosa</i> |
| 2. <i>Agrimonia gryposepala</i> | 18. <i>Carex intumescens</i> |
| 3. <i>Agrostis alba</i> | 19. <i>Carex normalis</i> |
| 4. <i>Agrostis perennans</i> | 20. <i>Carex rosea</i> |
| 5. <i>Alsine media</i> | 21. <i>Carex varia</i> |
| 6. <i>Anthemis Cotula</i> | 22. <i>Cerastium viscosum</i> |
| 7. <i>Aquilegia canadensis</i> | 23. <i>Chelone Lyoni</i> |
| 8. <i>Arctium lappa</i> | 24. <i>Chrysanthemum leucanthemum</i> |
| 9. <i>Aster divaricatus</i> | 25. <i>Cirsium discolor</i> |
| 10. <i>Aster patens</i> | 26. <i>Corylus rostrata</i> |
| 11. <i>Aster oblongifolius</i> | 27. <i>Crataegus iracunda</i> |
| 12. <i>Aster surculosus</i> | 28. <i>Dactylis glomerata</i> |
| 13. <i>Aster Tradescanti</i> | 29. <i>Danthonia compressa</i> |
| 14. <i>Azalea lutea</i> | 30. <i>Dennstaedtia punctilobula</i> |
| 15. <i>Carex communis</i> | 31. <i>Deschampsia flexuosa</i> |
| 16. <i>Carex crinita</i> | 32. <i>Diervilla sessilifolia</i> |

33. *Dracocephalum virginianum*
34. *Drosera rotundifolia*
35. *Eragrostis spectabilis*
36. *Erigeron ramosus*
37. *Eupatorium urticaefolium*
38. *Fragaria virginiana*
39. *Gaylussacia orecola*
40. *Gentiana quinquefolia*
41. *Glyceria melicaria*
42. *Gnaphalium purpureum*
43. *Grossularia rotundifolia*
44. *Helianthus decapetalus*
45. *Hieracium aurantiacum*
46. *Hieracium scabrum*
47. *Houstonia purpurea*
48. *Houstonia serpyllifolia*
49. *Hypericum Buckleyi*
50. *Hypericum punctatum*
51. *Hypericum virgatum*
52. *Ionactis lineariifolius*
53. *Juncus marginatus*
54. *Juncus tenuis*
55. *Kalmia latifolia*
56. *Kneiffia fruticosa*
57. *Koellia dubia*
58. *Koellia pycnanthemoides*
59. *Krigia virginica*
60. *Lechea Leggettii*
61. *Leontodon Taraxacum*
62. *Leptilon canadense*
63. *Lilium philadelphicum*
64. *Lilium superbum*
65. *Lobelia inflata*
66. *Lysimachia quadrifolia*
67. *Mesadenia triplifolia*
68. *Monarda clinopodia*
69. *Monarda media*
70. *Nepeta Cataria*
71. *Nothoholcus lanatus*
72. *Oxalis stricta*
73. *Oxypolis rigidus*
74. *Panicum meridionale*
75. *Pedicularis canadensis*
76. *Penstemon brevisepalus*
77. *Phlox ovata*
78. *Phleum pratense*
79. *Pieris floribunda*
80. *Plantago cordata*
81. *Plantago lanceolata*
82. *Plantago major*
83. *Poa compressa*
84. *Poa cuspidata*
85. *Poa pratensis*
86. *Polygonum aviculare*
87. *Polygonum persicaria*
88. *Polytrichum commune*
89. *Potentilla canadensis*
90. *Potentilla recta*
91. *Prunella vulgaris*
92. *Pteridium latiusculum*
93. *Rhododendron catawbiense*
94. *Rhododendron punctatum*
95. *Rubus allegheniensis*
96. *Rubus canadensis*
97. *Rudbeckia hirta*
98. *Rudbeckia laciniata*
99. *Rumex acetosella*
100. *Rumex obtusifolius*
101. *Salix humilis*
102. *Salix tristis*
103. *Schizachyrium scoparium*
104. *Senecio Smallii*
105. *Sibbaldiopsis tridentata*
106. *Silene virginica*
107. *Solidago bicolor*
108. *Solidago caesia*
109. *Solidago hispida*
110. *Solidago monticola*
111. *Solidago nemoralis*
112. *Solidago patula*
113. *Solidago speciosa*
114. *Stachys aspera*
115. *Specularia perfoliata*
116. *Trifolium pratense*
117. *Trifolium repens*
118. *Vaccinium corymbosum*
119. *Vaccinium hirsutum*
120. *Verbascum Thapsus*
121. *Veronica officinalis*
122. *Viburnum cassinoides*
123. *Viola lanceolata*
124. *Viola sororia*
125. *Xolisma ligustrina*

TRAIL BALD. Tennessee Ridge. 6000 ft. July, 1936. Along many hundred yards of the high trail on the boundary line between Haywood and Transylvania counties. With the mountain oat grass (*Danthonia compressa*) strongly dominant on and along the sides of the trail for extended distances, this trail bald together with many others like it, furnishes evidence for the human origin of the larger and broader balds. The grass originally appearing in the trail seems to be slowly encroaching on the narrow shrub borders transitional to the forest. So thick is the grass on the sloping sections of the ridge top trail that complete protection from gullying has been given.

COLD MT. BALD. Haywood County, N. C. 5900 ft. 14 acres. Sept., 1935. Extends along south side ridge top near and on east side of peak; includes a gap. 1—*Danthonia compressa*; 2—*Potentilla canadensis*; 3—*Achillea millefolium*; 4—9, 12, 38, 40, 47, 60, 51, 57, 72, 104, 105, 107; 5—24, 36, 92, 97, 109, 120. Bounded below by shrub bald zone (*Vaccinium corymbosum* dominant), transitional to *Quercus borealis* forest. Above just over ridge top *Crataegus* sp., dominant *Quercus borealis* and at the gap *Fagus grandifolia*. Evidence of fire in last 5 years. Spring near bald, lower side.

An old mountaineer met on this bald stated that he could discern no noticeable change in the relative size of the grass and shrub areas in the 50 years he had been visiting the bald for blueberries.

Cold Mt. with its twin peak, Shining Rock, both having balds was called "Datsunalasgunyi" by the Cherokees.

ANDREWS BALD. Smoky Mt. National Park, Swain Co., N. C. (Fig. 2). Roughly square in outline, 5860 ft. Reported as 75 acres. Probably an overestimate. July, 1925. On gentle southwest slope at end of high ridge 2 miles south of Clingman's Dome. An extraordinarily large and fine example of the ancient herbaceous balds originated by the Indians. Strongly dominated by species of *Carex* with the emphasis on the upper and west sides shifting to the single species *Carex flexuosa* because of higher soil water. The sedge cover on this bald was the thickest of all seen; the sod very deep and heavy. Plants of the middle and lower area: 2—*Carex varia*, *C. radiata*; 3—*Carex flexuosa*, *Potentilla canadensis*; 4—9, 16, 19, 20, 22, 29, 35, 38, 41, 64, 99, 112, 124; 5—30, 47, 48, 49, 73, 78, 84, 91, 93, 122. In an earlier note (19) too great an emphasis was given to the oat-grass; this bald is fundamentally a sedge bald.

This expansive area is dotted with a few widely scattered relict shrubs, *Viburnum cassinoides* and *Ribes rotundifolium* being prominent. East

side bounded by a tree covered declivity near ridge top: north side by balsam-spruce climax with *Rhododendron catawbiense* along the ecotone: west side balsam-spruce with mixed shrub border grading into low *Rubus*: South side balsam-spruce with *Glyceria* and heavy *Carex* at one part indicating an almost marshy character of the bald here. Good spring near southwest corner of the bald. No evidence of recent fire or grazing.

MT. STERLING BALD. Haywood Co., N. C. (Fig. 3). 5800 ft. Small rectangular bald 100 x 350 ft. lying on gentle western slope near top of the mountain, long axis parallel to contour lines. June, 1935. Notable for the high dominance of *Carex flexuosa* to be correlated with its wet soil character due to drainage from the balsam-spruce above. The central body of the bald shows: 1—*Carex flexuosa*; 2—none; 3—*Potentilla canadensis*, *Viola* sp., *Fragaria americana*; 4—18, 19, 22, 83, 95; 5—14, 28, 67, 78, 104. On the margin sociies of *Gaultheria procumbens*, *Dennstaedtia punctilobula*, and *Polytrichum* sp. were noted, also *Carex lucorum*. This bald is completely surrounded by the balsam-spruce climax. A spring is located a few yards from the lower trail entrance.

A bare area (Fig. 4) made on this bald on the tent site of Dr. H. L. Blomquist of Duke University during the summer of 1933 shows little seral progress. *Rumex acetosella* had appeared forming a marginal ring; the center was bare after two years.

ROAN BALD, N. C. Tennessee line near Bakersville, N. C. (Fig. 5) 5800 ft. Extends along ridge east of Carvers Gap, at present involving many hundred acres (most estimates 1000). Not visited since 1926. The relative dominance was not noted at that time. The common plants were *Danthonia compressa*, *Poa compressa*, *Trifolium repens*, *Potentilla canadensis*, *Houstonia serpyllifolia*, *Polytrichum commune*, *Veronica officinalis*, *Rumex acetosella*, *Deschampsia flexuosa*, *Agrostis alba*.

This is probably the largest high mountain grass area in the Southern Appalachians. All evidence indicates, however, that it owes its present large size to the activities of the early white mountaineers who destroyed large areas of forest for grazing purposes. The high North Carolina—Tennessee ridge at this point is unusually broad and flattened, making an extensive grazing area possible here. Relict buck-eyes (*Aesculus octandra*) were scattered on the lower slopes (Fig. 6). The limited area on the nearly flat top was the probable site of the original Indian bald. Here the *Danthonia* grass makes a heavy sod and the original bald must have closely resembled a bald like Gregory which

occupies a similar site. On the south slope the uncommon *Alnus* *Alnobetula* forms a thicket community. The surrounding forest border is of hard-woods of which beech and northern red oak are prominent.

This bald and its nearby forested peak (now cut over) were visited by the early taxonomists—Gray, Lamson-Scribner (13), Curtis, Buckley, and Chickering (5), none of whom discussed the probable origin and maintenance of such a grass area.

Lanman (14) reports an Indian tradition which tells of 3 great battles which were fought on this mountain, the Catawbans winning against the Cherokees. After these battles the Great Spirit caused "the forest to wither from the 3 peaks." Such a legend is an indication of the great antiquity of the bald.

SHINING ROCK GAP BALD. Haywood Co., N. C. (Fig. 7) 5700 ft. July, 1935. Two acres in the gap and extending up lower southwest slope. Gives way to shrubs on upper half of slope except for 2 small pure grass areas (1) a local level place, 30 ft. in dia. and (2) an acre-size area on the mountain top. Gap bald, 1—*Danthonia compressa*; 3—*Houstonia serpyllifolia*, *Vaccinium corymbosum* (relicts). 4—4, 32 (marginal), 47, 56, 89, 93, 95 (marginal), 99; 5—17, 66 (marginal), 71, 77. The upper two local balds are in almost pure mountain oatgrass broken by shrub relicts. The shrub bald ecotone to the forest is of an open type on the higher more gentle slope and massed on the lower steeper south-facing slope. *Kalmia latifolia* and *Rhododendron catawbiense* are the most abundant with *Pieris floribunda* represented. Beyond the shrubs is *Quercus borealis* and in a few places the shrub bald makes contact with the vast successional community of *Prunus pennsylvanicum*. On the higher side the grass and shrub areas are bounded by the quartz rock wall dike which gives the name to this peak. An excellent spring is located very near to the gap bald.

The shrubs of the upper slope and top are chiefly the blueberry (*Vaccinium corymbosum*) making this mountain one of the largest "berrying grounds" in the region (Fig. 8). The open character of the shrubs is due to this activity, the interlocking paths having grown up in oatgrass, a fact extremely significant in the problem of bald origin.

BUNCHES BALD. Haywood Co. near Soco Gap. 5700 ft. A small rectangular bald of two acres at the ridge top on the south slope. Notable for the presence in it of the northern *Hieracium aurantiacum*. 1—*Danthonia compressa* broken sharply in various small socies of fork dominants; 2—*Achillea millefolium* (socies); 3—*Erigeron ramosus*, *Potentilla canadensis*, low *Rubus* sp., *Chrysanthemum leucanthemum*; 4—

3, 22, 38, 45, 47, 71, 75, 76, 91, 99; 5—7, 60, 88, 104, 106, 117. The weedy character of this bald indicates recent disturbance in relation to the lumbering going on in the vicinity.

Bounded by a marginal shrub-community of *Vaccinium corymbosum*, *Lyonia ligustrina* and *Azalea calendulacea*. Also *Pteridium latiusculum* prominent. Back of the shrubs the *Quercus borealis* consociation surrounds the bald.

WEST ROUGH BUTT BALD. Boundary line between Haywood and Jackson counties. 5700 ft. June, 1936. Two almost contiguous areas of 5 acres each on west sloping ridge leading down from Rough Butt Mt. Lower bald of the two on a broad, gently rounded ridge top. Dominated by an almost pure stand of *Poa compressa*. 2—*Houstonia serpyllifolia*; 3—*Potentilla canadensis*. Bounded by mixed *Picea rubens* and *Quercus borealis*. Indications of heavy grazing.

TENNESSEE BALD. At junction of boundary lines of Haywood, Transylvania, and Jackson counties. June, 1936. 5622 ft. Six acres. A narrow rectangular bald occupying gap extended along gap ridge top lying on the long grade east slope, immediately below ridge top. 1—*Poa compressa*; 3—*Potentilla canadensis*, *Houstonia serpyllifolia*, *Juncus tenuis*; 4—1, 99, 121. The upper $\frac{1}{4}$ — $\frac{1}{2}$ of the treeless area is in shrubs with *Vaccinium corymbosum* dominant. Upper boundary *Quercus borealis* with some *Picea rubens*; the lower boundary *Betula lutea* with a few *Abies Fraseri*, *Prunus serotina* and *Acer spicatum*. An excellent spring is found at the middle of the bald, a few yards into the forest.

This gap bald was a well known one in the early days. A main trail crossed it and today as indicated above it is the junction of three counties. The Cherokees called it "Tsunegunyi" and on it and its neighboring balds lived the great legendary hornet (or bird in another account) whose activities were described earlier.

SILER'S BALD. N. C.—Tenn. line, Smoky Mt. Park seven miles west of Clingman's Dome. Aug., 1936. 5600 ft. Five acres on south spur of Siler's Bald Mt. beginning at mountain peak. This bald must be dealt with in three sections:

East Section: The ridge top in *Danthonia compressa* with sociies of *Potentilla canadensis*. The northwest slope below top in tall weeds and *Rubus*. Stumps of small beeches here showed recent cutting and fire.

Middle Section: Apparently was originally the main bald. The south slope at present in *Potentilla* and *Fragaria*, a ruderal society coming in after the *Danthonia* grass had been killed by the early 1936

drought. North slope dominated by *Solidago* with socies of *Helianthus*. On the moist lower slope of the middle section *Carex flexuosa* dominates with much *Rubus* under three feet.

West Section: Upper part at present chiefly in weeds, *Rudbeckia laciniata*, *Fragaria americana*, and *Potentilla canadensis*. Only two acres at base of bald with living *Danthonia compressa*.

This bald among many others was severely affected by the spring drouth of 1936 changing the high dominance of the oatgrass over to a mixed weed community.

HIGH SPRING BALD. N. C.—Tenn. line. Smoky Mountain Park. Five and a half miles west of Clingman's Dome. One-fourth acre. August, 1936. 5500 ft. A small bald to be correlated with a good spring on south side just below the ridge top. At present largely in weeds with *Phleum* prominent indicating recent human interference. The high spring found here is to be correlated with the water indicating *Carex* at east end, where subsoil drainage from neighboring ridge top passes on way to spring.

THUNDERHEAD MT. BALD. N. C.—Tenn. line, Smoky Mt. Park, near top of mountain. 5500 ft. Ten acres. July, 1936. This grass bald is exceptional in its location on a rather steep slope and that slope has a northwestern exposure. *Danthonia compressa* was in complete control and had been able because of the northern exposure of the site, to survive the 1936 spring drouth. It is bounded on its east side by a compact mass of *Rhododendron maximum* which extends up slope to the top of peak. A small oat-grass community 20 ft. in diameter covers the mountain summit. The western boundary is the high mountain hardwood forest dominated by *Quercus borealis*.

SOCO BALD. Haywood County, three miles from Soco Gap. July, 1935. 5400 ft. One and a half acres on south rather steep slope just below top of Soco Bald Mt. A mixed bald with low shrubs scattered through grass areas. On a relative dominance basis the record gives: 2—*Danthonia compressa*, *Vaccinium corymbosum*; 3—*Senecio aureus*; 4—*Lyonia ligustrina*. *Salix humilis*, *Erigeron ramosus*, *Pedicularis canadensis*, *Sassafras sassafras* (noteworthy), *Kalmia latifolia*, *Azalea calendulacea*. On the upper or ridge top side the bald is bounded by low *Fagus grandifolia*, *Azalea calendulacea*, and low *Castanea dentata*. Below, *Quercus borealis*.

This bald is introduced as a type of shrub bald containing enough grass to make its classification difficult. It is not a grass bald proper but represents a fire repressed shrub bald with a grass admixture.

BLOCK HOUSE MT. BALD. Near N. C.—Tenn. line. Two miles from

Thunderhead Mt., Smoky Mt. Park. July, 1936. 5400 ft. Three acres of bald proper (without trees). One on north slope of peak and two on south slope. On the upper $\frac{1}{4}$ of the latter the *Danthonia compressa* grass had been killed by the 1936 spring drouth and the area taken by weeds which in progressively decreasing size below gave way to the still living grass below. The north bald pure stand of oatgrass was unaffected by the drouth. The relative dominance of the weeds occupying the grass killed area was as follows: 2—*Solidago patula*, *Potentilla canadensis*; 3—*Rumex acetosella*, *R. obtusifolius*, *Aster* sp.; 4—*Phleum pratense*. The south bald is bounded on west by scrub *Fagus grandifolia* 1-4 ft. high on lower and east side by *Quercus borealis*. The north bald is entirely surrounded by beech.

Of especial interest is to be found on the forested mountain slope below the bald a great development of oatgrass under the trees extending down slope on the south side to Haw Gap, involving an area of 25 acres or more. Such an extensive grove of large old trees with almost pure grass beneath is very unusual in the high mountains. Its occurrence here may be ascribed to a combination of grazing and fire within historical times. The treeless areas above are regarded as true grass bald initiated by the Indians for game lures.

BIG PISGAH RIDGE BALD. Haywood County line ridge near Mt. Pisgah. 5340 ft. Aug., 1935. Two acres. A grass bald bordered by *Corylus rostrata*. Of rectangular shape, its long axis is on the ridge top, the south slope exposure being slightly larger than the north. 1—*Danthonia compressa*; 2—*Potentilla canadensis*; 3—*Phleum pratense*, *Pteridium latiusculum*; 4—24, 25, 57; 5—36, 66, 72, 91, 97, 99, 108, 112, 116, 118, (relicts), 125 (relicts).

Of especial interest on this and the following bald is the hazelnut (*Corylus rostrata*) border (Fig. 9) with a minor amount of *Salix humilis* on the south side. On the north side this shrub is massed in a pure stand between the *Quercus borealis* forest and the grass and by short rhizomes is slowly encroaching on the grass area. The west end of the bald opening is already choked. This bald is unquestionably being transformed into a *Corylus* shrub bald. The environing forest is that of *Quercus borealis*. Probably a lure bald as to origin; no spring was found near it.

LITTLE PISGAH RIDGE BALD. Near Haywood County line ridge, vicinity of Mt. Pisgah. 5330 ft. Sept., 1935. Only two small grass areas left amid the heavy *Corylus*, each roughly circular with diameters of 6 and 25 ft. respectively. The original bald opening was 8 acres.

1—*Danthonia compressa*; 3—*Potentilla canadensis*, *Pteridium latiusculum*; 4—*Cirsium discolor*, 5—*Prunella vulgaris*. This grass bald has been almost eliminated by the hazelnut, and gives an exact index as to the facts of the preceding. These two balds, now in transition from grass to shrub (*Corylus*), are the only ones of the kind noted. *Quercus borealis* surrounds the thick hazelnut community with some *Kalmia* and *Crataegus*.

INDIAN GRAVEYARD BALD. On south slope of a low mountain near Tennessee Ridge at headwaters of Pigeon River. 5300 ft. Aug., 1935. Four acres, roughly square in outline. At time visited the bald was dominated by *Solidago patula*, a successional stage following an earlier drouth. Scattered over this bald and in the surrounding forest as well were the mounds and depressions of a former extensive wind-fall. The mound axes are all parallel to the contour lines suggesting an artificial arrangement explained as "Indian graves" but in reality due to the uniformity of tornado wind direction. *Danthonia compressa* was developing between the masses of *Solidago* and was dominant on the mound tops. In addition to shrub relicts of *Cornus asperifolia* and *Vaccinium corymbosum* a few repressed trees of *Robinia pseudo-acacia* are presented, their presence made possible here by the relatively low altitude.

On the west and south margins the bald is in contact with a vast area of *Rubus* and *Prunus pennsylvanicum* with no plants of these present in the bald. On the east border is found *Fagus* and *Cornus*, the contact sharp. On the upper or south side, *Quercus borealis* was dominant.

LITTLE WAYAH BALD, Macon Co., near top of peak east of Nantahala Gap. July, 1935. 5000 ft. One-half acre on the southeast slope. Lower three-fourths of the opening in grass. 1—*Danthonia compressa*; 3—*Potentilla canadensis*, *Fragaria americana*; 4—22, 36, 58, 77, 91, 104; 5—1, 24, 60, 66, 71, 74, 95, 106, 120. The upper fourth of the forest opening is in shrubs of which the east half is in *Salix humilis*, west half *Kalmia* and *Azalea calendulacea*. The bald is bounded by *Quercus borealis* with some *Fagus* especially on the west side. No spring near by. Apparently used as a lookout because of the proximity to the gap close by.

SPENCE (OR SPENCER) FIELD BALD, N. C.—Tenn. line, Smoky Mt. National Park near Thunderhead Mt., lying between the headwaters of the west fork of the Little River and Gunna Creek. July, 1936. A long undulating ridge top bald a mile long and involving at least 40 acres, including the broad main gap. *Danthonia compressa* normally

highly dominant but large areas of it on the south slope near the ridge top were killed by the 1936 spring drouth, being replaced by 1—*Potentilla canadensis* and 4—*Prunella vulgaris*. On the north slope the oatgrass was highly dominant 1—*Danthonia compressa*; 3—*Solidago spatula*; 4—24, 99, 104, 121. An excellent spring is found near the gap on the south side. The south boundary consists of *Quercus borealis*, *Aesculus octandra*, and *Fagus grandifolia*. On the north side *Aesculus* is dominant with some *Sorbus americana* present. To the east on the ridge, relict trees of *Acer saccharum*. *Quercus alba* and *Betula lutea* are present in all stages of degeneration, under the combined attack of the axe, fire, grazing, and drouth.

This bald has unquestionably been enlarged in modern times through the heavy grazing imposed upon it by the mountaineers. Its gap nucleus, however, dates back to the Indian days.

CRABTREE BALD. Haywood Co., near Crabtree, N. C. (Fig. 10). July, 1935. 5280 ft. A recently developed bald of 20 acres on north slope of mountain from top down to the gap. 1—*Poa pratense*; 2—*Agrostis stolonifera*, *Achillea millifolium*; 3—*Danthonia compressa*, *Oxalis stricta*, *Cerastium viscosum*, *Phleum pratense*, *Trifolium repens*, *Potentilla canadensis*, *Cirsium odoratum*; 4—8, 28, 53, 99, 100; 5—78, 80, 90.

Scattered over this bald were relict specimens of *Robinia pseudo-acacia* in all stages of decay. Most of the trees were dead. One large mass of *Corylus rostrata* was noted near center and a few masses of *Dennstaedtia* fern. Above at the mountain top, the boundary forest was *Fagus* with *Aesculus octandra* relicts. Below the bald merged into a ridge meadow with abundant *Chrysanthemum leucanthemum*.

This bald is entirely of recent origin and is introduced here as an example of a bald made by the white pioneers who were able to extend their grazing areas with fire aiding their axes. Such balds are, however, not common.

BIG CHESTNUT BALD. On N. C.—Tenn. line peak in the Smoky Mt. Park at the head of Defeat Branch of Valley Creek. Sept., 1935. 4970 ft. Forty-five acres. An irregularly shaped ridge bald lying chiefly on the south slope. Much disturbed in recent decades and invaded by weeds and blackberry. At the east end lies a restricted area of typical oatgrass bald. 1—*Danthonia compressa*; 3—*Potentilla canadensis*; 4—66, 107, *Aster* sp., *Solidago* sp.; 5—64, *Lactuca* sp. One large *Acer saccharum* tree stands in this grass area.

In the vicinity of the ruins (chiefly a fallen-in chimney) of the old

Hall's cabin was a ruderal community as follows: 1—*Phleum*; 3—*Fragaria americana*, *Aster tradescanti*, *Rudbeckia laciniata*; 4—1, 2, 8, 72, 100.

This bald opening is surrounded by *Quercus borealis* trees which at the very top of the mountain above the bald reach the unusual height of 40–50 ft. in spite of wind exposure. *Castanea* and *Aesculus* also present.

GREGORY BALD. N. C.—Tenn. Line (Fig. 11). Smoky Mt. Park near west end of Park. Aug., 1935. 4948 ft. Seventy-five acres. This bald is one of the largest known. Roughly rectangular in shape, it is located on an unusually flat high prominence of the main ridge. It extends down on the north side but 20–50 yards while on the south side to fully 150–160 yards. 1—*Danthonia compressa*; 2—*Juncus tenuis*; 3—*Muhlenbergia sobolifera*; 4—22, 99, 102; 5—11, 24, 36, 42, 47, 48, 65, 71, 72, 104, 115, 116, 117, 120, relicts of 14, 125, *Vaccinium hirsutum* and *Aronia nigra*.

In the central region at the top is a rocky area of $\frac{1}{2}$ acre with a weed community as follows: 1—*Trifolium repens*; 2—*Polygonum persicaria*; 3—*Polygonum aviculare*, *Juncus tenuis*, *Rumex acetosella*. The north boundary is of scrub *Quercus borealis* with a transition zone of shrubs (*Lyonia ligustrina*, *Vaccinium corymbosum* and others). The south tree boundary is also of *Quercus borealis*, but higher trees in front of which is a broad transition zone of mixed shrubs. Especially noteworthy here was the occurrence in large masses of the northern shrub willow *Salix tristis* which made the contact with the grass while *Vaccinium corymbosum* was next to the forest.

This bald was famous among the Cherokees as being "The rabbit place" (Tsistuyi). Here the chief of the rabbits lived. He was as large as a deer and ruled all of the rabbits of the Cherokee country.

Camp (3) holds "that the area was not originally a pure grassy meadow but one with numerous shrub islands of various types and mentions a tradition that this bald was "originally a blueberry meadow." He thus believes it was "once dominantly a heath bald containing open spaces with a heavy covering of grasses, sedges, and other herbaceous plants." He (4) later rightly states that it "bears on its great wind swept dome the most striking and choice assortment of ericaceous genera and species of any of the grassy balds." He found the hybridizing of at least 3 species of *Azalea* result in a polyglot population almost impossible to unravel.

OLD CAMP BALD. In gap 300 feet below Little Wayah Bald, near

Nantahala Gap. July, 1935. 4700 ft. One and a half acres. The undisturbed $\frac{2}{3}$ of this bald was dominated by oatgrass and closely similar to Little Wayah Bald nearby. Of especial interest was the composition of a semi-weedy disturbed area, the site of an old cabin and corral. 2—*Salix humilis*; 3—*Fragaria americana*, *Solidago* sp., *Danthonia compressa*; 4—*Prunella vulgaris*; 5—*Diervilla sessilifolia*. The bald opening was closely surrounded by a *Quercus borealis* forest.

The significant fact here was that of the reinvasion of the oatgrass into the weed community. Every indication pointed to the gradual restoration of dominance to this plant.

GENERAL DISCUSSION

The observation that typical bald communities had appeared on many hundreds of yards of the high ridge trails, was the basis for the idea that the prehistoric grass balds must have had a similar origin to the perfectly evident origin of the unrecognized linear balds of the trails. These trail balds, of which examples on Tennessee Ridge and Shining Rock Mt. (Fig. 12) have been described, could have had no other origin than that of the succession which took place after the trail became sufficiently unused to make possible the appearance in it of an herbaceous community. Fire conducting itself along a ridge-top and confining itself to the trail and immediate trail border is of course an absurdity.

The pioneer dominants must have been herbaceous for in many places on sloping ridge tops these bald trails are sunken, indicating a degree of use and weather erosion which eliminated the reproductive parts of woody plants, rhizomes, stolons, and seedlings. Following a brief weed-like pioneer stage initiated commonly by *Rumex acetosella*, these trail areas went over to a high dominance of the mountain oatgrass (*Danthonia compressa*). In many places, this remarkable grass seems to have extended its range laterally at the expense of the narrow zone of shrubs which commonly is found transitional to the all-environmenting forest.

From the trail bald it is a simple logical step to realize that the broader local areas recognized as "grass balds" must have had a similar origin though severe and intensive local human interference which as will be shown later, must occur, to initiate an herbaceous succession in contrast to the usual woody succession following fire or lumbering. These ancient grass balds such as Andrews and Siler's in the Smokies and Cold Mt. and Shining Rock in the Balsams are then essentially

expanded trails. Antidating, as most of them do, the coming of the white man, they unquestionably owe their origin to sufficient intensive local and continuous destruction of the woody vegetation to make possible the advent of an herbaceous complex, progressing rapidly to the stable *Danthonia* community on mesic soil habitats or *Carex* on wet ones. Having attained dominance these perennials with and apparently without the aid of fire are able to compete successfully against the woody plants.

Camp's (3) data on Gregory Bald (given previously) strongly supports this theory, though he does not use it as it is interpreted here. His movement from shrub to grass is due he believes to "hot south-westerly winds."

INDIAN CAMP SITES

It may now be clearly understood why 15 of the 22 old balds described herein are found on gentle warm south-facing slopes of high ridge tops, gaps or knobs, just above or near to springs. These are interpreted as summer time camp sites where the Indians may have gathered in large numbers for it is known that they preferred the ridge trails for hunting and travel over the stream side ones. Truett (16), a student of trade and travel before 1830, states:

"In the mountain and hill country the trails usually led along higher ground and ridges where the undergrowth was not so dense and where there were fewer streams to cross. The trails along ridges also afforded good opportunity for sighting game and enemies. These two factors determine to a great extent the location for paths."

The Southern Appalachian ridges are so massive that ascent and descent on the same day leaves little time for hunting or any other activity including ecological note taking (as the writer repeatedly discovered), hence the development of local high mountain camp sites would prove a real need for the Indian hunters. It is entirely probable that during the summer the women and children for varying periods would be taken to such camp areas. Further the large balds may have been refuges for large numbers of Indians in time of war.

Some of the balds which in the mind of the writer were originally major Indian summertime camp sites are: Shining Rock, Andrews, Tennessee, Siler's, Spence Field (near Thunderhead Mt.), Big Chestnut, Gregory, Roan. There are numerous minor grass balds such as High Spring Bald earlier described which were unimportant as a camp site but were significant as a meeting ground at the highly placed spring.

It must be stated that springs located near ridge tops are not frequent in the Southern Appalachian Mountains. In the eastern and more rugged half of the Smoky Mt. Park there are only a few places where water may be had convenient to the high trails.

The sharp angular character of the boundary of certain balds such as may be seen at Andrew's and Mt. Sterling balds, has a highly artificial appearance. In both of these, sharp right angles occur in the line of contact between balsam trees and the grass. No set of natural factors would be so arbitrary in a boundary line.

In contrast, a most important early trail crossing; viz., Indian Gap on the main Smoky ridge over which passed one of the great Indian war trails formerly showed but a few square yards of grass developed at the sharp ridge top. And no water is to be found within many hundred feet.

As to the time these camps were occupied nothing is known from the early records. The "between the lines" inference in the Cherokee legends is that the balds far antedated the 18th Century Cherokee. Their actual origin probably goes back many centuries before the advent of the white man and they may be relicts of a culture and of habits, about which the Cherokee of the DeSoto and Bartram periods knew nothing.

GAME LURES

After publishing a preliminary note (18) on the Indian theory of grass bald origin, the writer learned from Mr. Arthur Stupka, Smoky Mt. Park naturalist, an additional line of evidence. Near the Mt. Sterling Bald (previously described) Mr. Stupka met an old mountaineer, who reported that his grandfather had been told by the Indians that the Mt. Sterling Bald had been made by the Indians for a game lure. Deer and wild turkeys would be tempted into the opening and be easily shot with bow and arrow from the deep shadows of the environing balsam forest.

This contribution is especially interesting in light of the fact that the Mt. Sterling Bald is a wet bald dominated by *Carex flexuosa* and would in wet seasons be unfit for camping purposes.

Another bald of this probable origin and purpose is one located on the slope of the ridge between Clingman's Dome and Siler's Bald which from its shape may be known as Square Bald. These game lure balds unadapted for camping are few in number.

LOOKOUTS

A third type of bald is located near or on sharp summits and which seem to have been little more than lookout stations. Such a one is the small bald at the top of Blockhouse Mt., a neighboring peak to Thunderhead Mt. in the Smoky Mt. Park, but not on the main high N. C.—Tenn. ridge. This bald faces to the nearby main ridge and also overlooks the Gunna Creek valley. It would be most reasonable to regard such a small bald as a signal station. Similar small balds near the top of such strategic peaks as Shining Rock, Cold, Bunches, and Wayah seem to find their proper classification in this category.

OBJECTIONS TO INDIAN THEORY ANSWERED

Only two students mention the Indian in connection with the balds, only to eliminate him from the picture.

Fink (8) believes they "could not have been cleared by the Indians in prehistoric days for it would have been too gigantic a task far beyond the possibility of their rude stone axes. Indian energy, or rather lack of it, would not have gone to such incredible labor without some vitally impelling cause. And even had they done so in the centuries which must have elapsed the clearing would have been completely reforested."

If we are careful to subtract the marginal areas on many of the balds which were made by the white mountaineer and his cattle and horses, we do not have a "gigantic" task to clear what is left. The original balds were unquestionably at first highly restricted expansions of the trails at these strategic points which through decades of use and slow destruction of trees and shrubs for fire wood expanded to the size at which the white mountaineer found them. The rude axe of the Indian plus many decades, or even centuries, of time could easily have enlarged the balds to the size first noted by the white men. As has already been shown, shrub bald areas may apparently be reduced to grass with the aid of criss-cross trails alone, though in many of the very open balds such as Andrew's, mass camping would be indicated to account for the extreme reduction of the woody plants. Such mass activity could easily have occurred in the summer period in relation to hunting or large war parties.

That under abandonment the balds would have been reforested, does not follow. All the evidence indicates that at the higher altitudes balds are completely resistant to invasion. The competition of the remarkable oatgrass is too severe for the woody plants. This grass

(*Danthonia compressa*) is confined to the high altitudes where on mesic sites, it competes so successfully with all other herbaceous plants as to reduce them to very minor elements of the population.

Davis (6) merely asserts that there is no "legend or evidence of Indian occupancy." This is a correct statement insofar as historical records go. The absence of such evidence in early records, however, may only mean that most early visitors to the Cherokee region never raised the problem of bald origin. Ecological concepts were of little concern to the early naturalists. There is little hope of getting any help on the problem from old records. But this lack does not detract to any degree from the purely ecological evidence presented in this study.

NEGATIVE EVIDENCE

The great single fact which constitutes strong negative evidence is that the subseres following fire, lumbering, or these two combined is initiated and developed by woody plants. At the higher altitudes in the balsam-spruce areas following the destruction of the forest the familiar fire cherry (*Prunus pennsylvanicum*) appears like magic from its long-viable seeds and by means of root crown shoots preserves its dominance under repeated fire. In the shade of this seral dominant, the balsam and spruce return. Often associated with this tree as sub-dominants are the bramble (*Rubus allegheniensis* and often the thornless *R. canadensis*) and the red elder (*Sambucus racemosus*).

This successional forest may be observed today covering many square miles of the high Balsams where it came in some 17 years ago following the removal of the forest with succeeding fire. And nowhere is there any evidence of any part of this vast deciduous successional cover giving way to a weed or a grass complex without the additional interference of man.

A similar relation holds at the slightly lower altitude or in the northern red oak (*Quercus borealis*) zone contiguous to the balsam-spruce with the difference that the bramble and other shrubs such as *Corylus* tend to gain control quickly and hold it until the trees are slowly replaced. •

Thus it may be stated categorically that if fire alone can produce a mountain grass bald, vast areas of the high Southern Appalachians would be in such balds. Yet the grass balds are but mere dots scattered widely over the vast mountainscape.

This important line of evidence together with the positive evidence

given earlier, constitutes strong support for the theory that the recognizably old or prehistoric grass balds are aberrant climaxes initiated through human interference (disclimaxes of Clements) which persist because of the highly competitive nature of the dominant grass in resisting encroachment of the woody vegetation at high altitudes.

It is worthy of record here that the dominant grass of the ecologically puzzling western high mountain deer parks, is a close relative of the eastern bald dominant; viz., *Danthonia californica* var. *unispicata*. Whether or not the theory developed in this paper could be made applicable to the western areas is for the consideration of those who know those openings better than does the author.

RECENT HUMAN INTERFERENCE

The influence of the white mountaineer on the balds has been through his use of these areas for grazing. In many cases by the use of his axe in addition to his stock, he has measurably enlarged the original balds. An excellent example of such a greatly enlarged bald is that on Roan Mountain which covers many hundred acres. It is interesting to note that those balds which show evidence on their margins of recent deforestation accompanied by grazing, are located at or near accessible, broad gaps, where the old mountaineer cross-ridge trails passed. Spence's Field near Thunderhead Mt. is another such bald enlarged and extended along the ridge by the mountaineer and his cattle.

More rarely a bald may be encountered which has been entirely initiated by the early native whites. Crabtree Bald, described earlier, is such a one. A few relict trees which survived the axe and fire are scattered over it in all stages of deterioration. Unlike nearly all of the larger balds, this one is on a north facing slope. The surest criterion, however, is the depth of the sod which in a bald of recent origin is notably thinner than in an ancient Indian initiated type such as Andrew's. The dominant Kentucky blue-grass, together with the relatively high dominance of a number of weeds, presents on this "bald" a completely different picture from that of the undisturbed true bald on which the mountain oatgrass or sedges are almost pure dominants.

RESPONSE TO GRAZING

The degree of grazing on the balds is most accurately indicated by *Juncus tenuis*, the path rush. On a heavily grazed and trampled area like that of Gregory, the rush attains a rank of 2 or that of a true sub-dominant with a marked suppression to a rank of 4 to 5 of the other

weed elements of the community. Andrews Bald shows no *Juncus* whatever. On the $\frac{1}{2}$ acre at the top of Gregory where the cattle gather at night with consequent eliminating of the grass, a destructive ruderal flora and dominance is to be noted.

With the cessation of grazing, no matter how severe the destruction of the grass, the seral progression is toward the *Danthonia* disclimax. However, short or long the intermediate weed-like stages are they are for the most part highly successful in holding the areas against the woody plants.

It is interesting to note that a few of the pioneer weeds are able to persist in the mature *Danthonia* or *Carex* communities in a highly suppressed state, ranking only 4 or 5.

On Old Camp Bald, described earlier, on the site of the former corral where the vegetation must have been practically eliminated, the oatgrass was returning and entering into competition with the wild strawberry and the *Salix humilis*, which had been able to establish itself on the disturbed area. How long the pioneers or later herbs other than the oatgrass hold these disturbed areas is not known. The Indian Graveyard Bald has apparently been for a number of years dominated by the perennial *Solidago canadensis* which gained ascendancy during a drouth year. The only places where the oatgrass was making a successful ecesis was on the mound summits. In time, the oatgrass would regain dominance over the area.

The Big and Little Pisgah Bald, which are rapidly being eliminated by *Corylus rostrata* moving into them from the margins by means of rhizomes, present a problem. Why have not all of the grass balds long ago been taken by the hazelnut? The answer is to be found, we believe, in two facts: (1) The low altitude of these balds making contact with the hazelnut climatic zone and (2) the heavy grazing these balds have had in recent years weakening the grass in its competition with this one shrub which was unmistakably and rapidly erasing the herbaceous community from the local mountain picture.

THE DROUTH CYCLE

A severe drouth occurred in the early summer of 1936 which resulted in the death of the oatgrass (*Danthonia compressa*) over relatively large areas of many of the balds. Only where a shower had fallen in time or where the soil depth or other soil factors had made possible the persistence of available water had the grass remained alive. The Smoky Mt. Park balds suffered more severely than did those in the Balsams.

The initiation of the subsera following the death of the dominant grass was carried out by a few species of weed-like plants (*Potentilla*, *Fragaria*, *Solidago*), which as highly suppressed perennials under the subsequent rains and the removal of the root competition rapidly developed into maximum sized plants. These seral communities of ruderal aspect in a few years give way again to the oatgrass and the disclimax grass community is restored until the next drouth occurs.

One very instructive situation was observed on Block House Mountain Bald. The grass near the ridge top had been killed *en masse* but lower down the slope it had survived the drouth. Over the killed

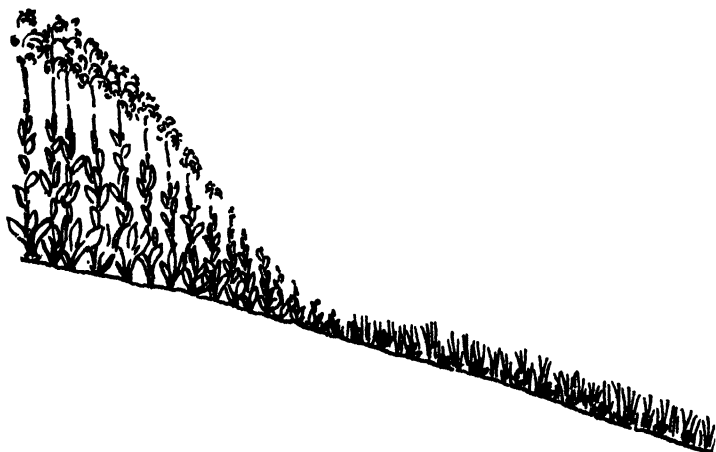


FIG. 13. DIAGRAMMATIC DEMONSTRATION OF INTENSITY OF COMPETITION SET UP BY THE OAT GRASS

At left, the response of the perennial goldenrod (*S. patula*) after the killing of the grass by drouth. Right, the repressed goldenrod in area of surviving grass.

area *Solidago patula* had assumed control but in the transition region to the living grass, the plants of this perennial species became progressively reduced in size until those which were hidden amid the luxuriant living grass became so suppressed by competition as to bear only two or three leaves (Fig. 13). This observation furnishes concrete evidence of the highly competitive nature of the oatgrass. When it is observed that these perennial goldenrods are kept almost completely suppressed it may be realized how impossible it would be for a seedling plant of any kind to become established and the explanation for the non-ecesis of the woody plants becomes apparent.

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SUMMARY

Twenty-three local grass or sedge covered areas ("grass balds") in the high Southern Appalachians are described and the theory put forward that they could have come into existence only through human activity, presumably that of the Cherokee Indians.

The Cherokee legends which indicate their presence in prehistoric times and the modern theories of scientists as to their origin (fire, ice storms, hot winds) and maintenance are given

The plant distribution is presented on a 1-5 scale of dominance, with an initial list of 125 species. *Danthonia compressa* (mountain oatgrass) is the most important species, being replaced as dominant in the occasional wet bald by *Carex* species, especially *Carex flexuosa*.

The Indian origin theory is based in part upon the negative evidence that every natural destructive agent (uncontrolled fire, insects, ice) initiates a woody plant succession and not an herbaceous one. Also during recent millenia any natural factor or complex of factors that could initiate and maintain herbaceous balds would have reduced extensive areas of the high mountains to this state.

The positive evidence is derived from the following facts: Most of the balds are on gentle south slopes of rounded ridge tops or gaps near high "bold" springs, indicating clearings for summer camp sites. A few are isolated on steeper slopes and have been reported (Mt. Sterling Bald) as having been made by the Indians for game lures and certain small ones on peak tops are to be interpreted as lookout points.

A most important line of evidence is that observable on many of the high ridge trails, where long sections have developed the bald plant complex, showing that the grass community may appear only following a long and severe disturbance of the original vegetation at the soil level. A grass bald is in a sense an expanded trail.

Following abandonment of these intensively disturbed areas, a ruderal or weed stage ensues, followed by the permanent grass (*Dan-*

thonia) or sedge (*Carex*) community. During drouth summers much of the grass may be killed and a new subserc initiated by the ruderal plants. The high mountain grass and sedge are able to compete successfully with the woody vegetation without the aid of fire, accounting for the maintenance of these artificial areas through centuries of time.

NORTH CAROLINA STATE COLLEGE,
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BIBLIOGRAPHY

1. CAIN, STANLEY. Ecological studies of the vegetation of the Great Smoky Mountains of North Carolina and Tennessee I. Bot. Gaz. **91**: 1-14. 1931.
2. . Ecological work on the Great Smoky Mt. region. Jour. So. Appal. Bot. Club **1**: 25-32. 1936.
3. CAMP, W. H. The grass balds of the Great Smoky Mountains of Tennessee and North Carolina. Ohio Jour. Sci. **31**: 157-164. 1931.
4. On Appalachian Trails. Jour. N. Y. Bot. Garden **37**: 249-265. 1936.
5. CHICKERING, J. W. Roan Mt. Bot. Gaz. **5**: 144. 1880.
6. DAVIS, JOHN H. Vegetation of the Black Mountains of North Carolina: An Ecological study. Elisha Mitchell Jour. **45**: 291-318. 1930.
7. EDSON, MRS. HELEN R. Frost forms on Roan Mt. Pop. Sci. Mo. **45**: 30. 1891.
8. FINK, PAUL M. A forest enigma. Am. Forests **37**: 538. 1931.
9. GRAY, ASA. Notes on a botanical excursion to the mountains of North Carolina. Am. Jour. Sci. & Arts. **42**: No. 1. 1842.
10. HARSBERGER, J. W. An ecological study of the flora of mountainous North Carolina. Bot. Gaz. **36**: 368-383. 1903.
11. . Phytogeography of N. America. New York. 1911.
12. JOHNSON, L. N. A tramp in the North Carolina Mountains. Bot. Gaz. **13**: 269 and 318. 1888.
13. LAMSON-SCHIBNER, F. The grasses of Roan Mt., Bot. Gaz. **14**: 253-255. 1889.
14. LANMAN, CHARLES. Adventures in the wilds of America. London, 1854.
15. MOONEY, JAS. The Cherokee Indians. 19th Rept. U. S. Bur. of Ethnology. 1898.
16. TRUETT, R. B. Trade and travel around the Southern Appalachians before 1830. N. C. Univ. Press. 1935.
17. WELLS, B. W. The Natural Gardens of North Carolina. Univ. of N. C. Press. 1932.
18. . Origin of the Southern Appalachian Grass Balds. Science **83**: 283. 1936.
19. WELLS, B. W. Andrews Bald: The Problem of its Origin. Jour. So. Appal. Bot. Club **1**: 59-62. 1936.

PLATE 1

- Fig. 2 (top). Andrews Bald. Note luxuriance of sedge cover, shrub relict and *Lilium superbum* at right.
- Fig. 3 (middle). Mt. Sterling Bald. Note sharpness of contact with the balsam forest.
- Fig. 4 (bottom). Camp site of Dr. H. L. Blomquist on Mt. Sterling Bald. Little revegetation in 2 years.

PLATE 2

- Fig. 5 (above). General view Roan Bald. Largely recent in origin.
- Fig. 6 (below). Roan Mt. Bald. Note relict buckeyes. Chiefly recent in origin.

PLATE 3

- Fig. 7 (above). Shining Rock Gap Bald. Note fire or pin cherry following logging and fire in the balsam area. No cherry whatever in the bald portion.
- Fig. 8 (below). Shining Rock Gap Bald (foreground). Shrub bald broken by blueberry pickers, grass developing in the trails.

PLATE 4

- Fig. 9 (above). Big Pisgah Bald. Note *Corylus* border encroaching laterally on grass area. *Cirsium discolor* along trail in foreground.
- Fig. 10 (below). Crabtree Bald. Recent in origin. Note relict trees.

PLATE 5

- Fig. 11 (above). Gregory Bald. Note sharp contact with northern red oak forest. *Salix tristis* in foreground.
- Fig. 12 (below). *Danthonia compressa* dominant in trail.

PLATE 1



FIG. 2

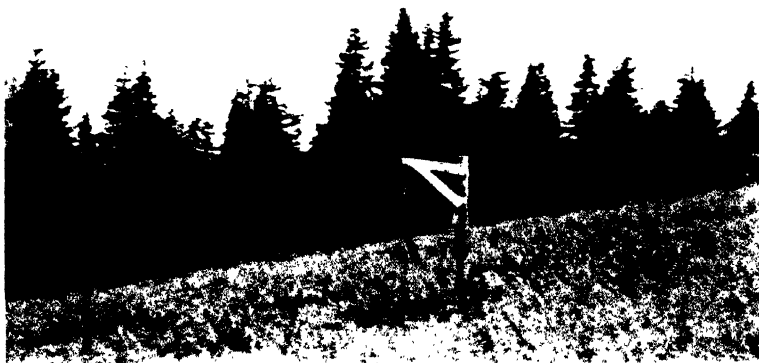


FIG. 3



FIG. 4

PLATE 2



FIG. 5



FIG. 6

PLATE 3



Fig 7



Fig 8

PLATE 4

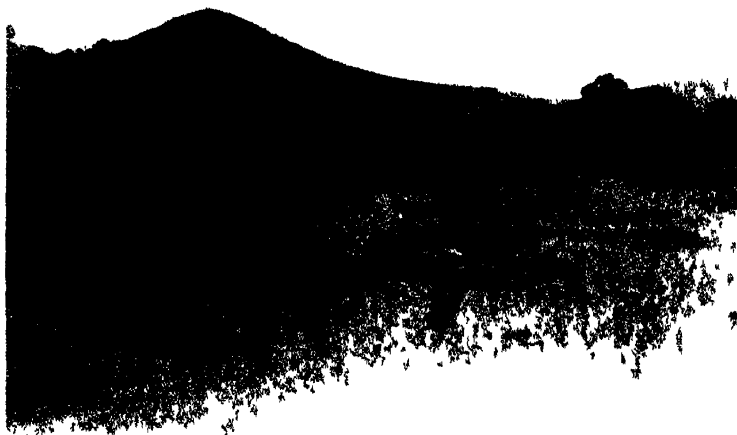


FIG. 9



FIG. 10

PLATE 5



Fig. 11



Fig. 12

NOTES ON MAYFLIES OF THE SOUTHEASTERN STATES (EPHEMEROPTERA)

By JAY R. TRAVER

PLATE 6

A six-weeks collecting trip in the southeast, * during the latter part of May and the entire month of June, 1936, yielded much material, an account of which is herewith presented. Seven new species are described. Notes on the life history and ecology of previously-known species, and descriptions of hitherto-unknown stages in the life history of others, comprise most of the remainder of the paper. Twenty-one species are added to the list of known North Carolina mayflies. Of the fifteen species reported from Alabama, fourteen are new records for the state. In addition, new records for the occurrence of certain species of mayflies in other southeastern states are presented

NORTH CAROLINA

Most of the collecting and rearing of specimens was done in the Appalachian region of North Carolina, and in the north-central portion of Alabama. In North Carolina, headquarters were established at Valle Crucis School, Valle Crucis (May 24-June 10), and at Penrose (June 11-June 24). From these points, many trips were made to adjacent areas. Nymphs collected elsewhere were brought back to headquarters for rearing. It was possible to keep rearing cages containing the nymphs in one of the small streams flowing through the valley, at Valle Crucis, and under a fortunate leak in a pipe line leading from a small mountain stream at Penrose. There were no casualties from floods during the past season's collecting, nor were the cages disturbed by too inquisitive humans.

In the Valle Crucis region in which collections were made, drainage is largely northwestward into the Watauga River; however, streams near Sugar Grove flow northward into the New River, while the North Toe and the Linville Rivers flow southwest and south respectively. Some

* This trip was made possible by reason of a grant from the American Association for the Advancement of Science.

of the species found in the Watauga River drainage are the same as those known to occur as far north as New York; others have been taken only in North Carolina or southward. While much new territory was covered on this collecting trip, many of the same streams were visited in which I had found good collecting in previous seasons.

In this connection, the condition of certain of the streams visited, as compared with those same streams in the summers of 1929 and 1930, may be of interest. Thus the Tuckaseegee River below Dillsboro is now practically devoid of all aquatic insect life, due to pollution of the stream by industrial wastes. On a pleasant warm June evening spent at Bryson City, through which town the river flows, not one stonefly, caddisfly, nor mayfly was to be found in the town or above the stream. One glance at the dirty, scum-covered black water was sufficient answer to our query as to their absence. In 1929, the stream still afforded fairly good collecting. Portions of the Davidson River, where gravel and sand are being excavated, yielded little or no aquatic life. The lower reaches of the North Fork of the Swannanoa River are reduced to a barren waste, due to silt pollution from similar excavating activities. The main stream of the Swannanoa was likewise a very poor collecting ground in 1936, although many specimens were taken there in previous years. The probable reason for this was not ascertained.

The entire area of the Great Smoky National Park is here considered under North Carolina, although certain of the specimens were taken from the Tennessee portion of the park. Likewise a few specimens taken at Trade, Tenn., just across the border from Sugar Grove, are listed under North Carolina instead of Tennessee.

Family EPHEMERIDAE

Subfamily Ephemerinae

Genus *Hexagenia* Walsh

**Hexagenia marilandica* Trav.

This species has not been reported previously from North Carolina. Nymph skins in abundance were found floating on the surface of the water near shore, at Lake Lanier, N. C. (This is a large artificial lake south of Tryon, almost on the North Carolina-South Carolina line.) A single male subimago was captured as it rested on a clump of reeds beside the shore, before taking flight. It failed to transform to the

* Species for which no references are given are treated fully in the *Biology of Mayflies*,—Needham, Traver, Hsu, 1935; Comstock Publishing Co. References for other species are indicated.

adult stage. No other adult specimens were obtained, in spite of an extensive search of trees, shrubs and low herbage near the lake shore.

Nymph (described from nymph sloughs). Body (including tusks): female, 40–42 mm.; male, 27 mm. Tusks slightly longer, more slender, and less sharply upturned than in the nymph of *H. carolina* Trav. (see fig. 16); frontal process of head of female more dome-shaped, not as conical as in the latter species. Head and thorax dark red-brown above; wing-pads deep blackish brown (somewhat darker than similar areas of *carolina*). Gills and gill-fringes deep purplish. Color and markings of body cannot be determined from nymph slough. In general, this nymph is slightly larger and stouter than *carolina*, a condition which is especially evident in the female.

Male subimago (dried). Body 21 mm.; wing 16 mm. Resembles *H. marilandica* in abdominal markings, in the very short distal joints of the forceps, and in the type of penes. In size, it is nearer to *H. rosacea* Trav., which species I had at first considered it to be. Since the single specimen is a subimago, the relative lengths of joints of the fore leg and the size of the eye cannot be compared satisfactorily with the types of either of these species. See record of this species also from South Carolina.

***Hexagenia kanuga* sp. nov.**

Male imago (specimen in alcohol). Body 20 mm.; wing 16½ mm. Head yellow. Median carina largely blackish; black spot at inner corner of eye, connected to black area on carina by a narrow black line. Ocelli black-ringed at base. Alabaster white areas on vertex and occiput, next to eyes and behind ocelli; narrow red-brown markings on these white patches. Upper portion of eye bright yellow; lower portion black. Pronotum yellow; two wide brown submedian stripes. Mesonotum yellow; red-brown markings along lateral margins and anterior to wing roots; scutellum greyish black. Metanotum dark brown in median area, yellow laterally. Pleura yellow; a dark brown triangle on basal portion of mesothorax, anterior to middle leg; an oblique streak extends upward from this triangle to wing roots. Sternum yellow. Dark brown triangles anterior to middle legs are joined by a median dark area, to form a continuous dark ventral band; from this, two submedian streaks extend backward almost to the metasternum. A purplish brown median patch on prosternum; metasternum largely dark brown. Fore legs missing, except trochanters, which are dark red-brown. Middle and hind legs yellow; claws and distal tarsal joints black, apex of preceding joint dusky. Wings hyaline. Costal strip of

fore wing stained with clear brown; pale at base, becoming more intense toward apex. A few cross veins in radial area basad of bulla are narrowly dark-margined; no large dark spots. Costa and subcosta brown; radius yellow-brown; other veins purplish black. In hind wing, outer margin purplish black in patches, the border thus discontinuous. Several cross veins widely dark-margined, so that 5 or 6 small dark spots are formed.

Abdomen yellow, marked dorsally with blackish brown, with red-brown ventrally. Tergites 1 and 2 largely dark; pale lateral margins and a pair of small pale submedian spots on each. A continuous dark median streak the length of the abdomen dorsally; gradually widened apically. Lateral oblique streaks run from this stripe forward to antero-lateral margin, on tergites 3-7. On 8 and 9, these streaks are shorter, ending in pleural fold halfway from base. On 10, they almost attain the anterior margin. Ventrally, red-brown triangles occupy most of the median area of each sternite. The apex of each triangle reaches only about halfway from base to anterior margin, but a narrow reddish mid-ventral line extends the length of the abdomen. A pair of pale, clear, submedian spots on each sternite, enclosed by dark triangle. On each sternite, next to pleural fold, a short, reddish, transverse dash. Genitalia of the *carolina* type. Forceps and penes yellow, streaked with purplish black; tips of forceps darkened. Distal joints of forceps as in Ulmer's figure* of typical *H. limbata* Guer. Tails largely yellow; beyond base, apical half of each joint may be brown; approximately every fourth joint is almost wholly brown.

This species *may* be synonymous with *limbata*, which is imperfectly known. However, a specimen in the Cornell collection, from Kansas, is much closer to the original description of *limbata* than is this specimen from North Carolina. The Kanuga Lake male differs from *limbata*: (1) meso- and meta-thorax yellowish instead of brown; (2) small but distinct dark spots are present in the hind wings; (3) brown markings of abdominal tergites are somewhat more extensive, especially on the basal segments. The ventral appearance of *limbata* is not indicated in the early descriptions, nor has it been described or figured in any more recent accounts.

Four male subimagos and four female imagos taken at Florence, S. C., seem to be of this species.

Female imago. Body 22-28 mm.; wing 20-22 mm. Body largely yellowish white. Longitudinal veins yellow, cross veins black, no dis-

* 1921—Ulmer, Georg. Ueber einige Ephemeropteren-typen älterer Autoren. Arch. f. Naturg. Abt. A, 6: 237, fig. 7.

tinct dark spots in wings; humeral cross vein infuscated in basal half. Basal abdominal tergites marked as in male, but paler. On all other tergites, a wide purplish black median stripe; faint traces only of the lateral oblique streaks, on tergites 3 and 4. Ventrally, a narrow black mid-ventral line on anterior half of each sternite is the only marking. Tails wholly yellowish.

Holotype—Male imago. Kanuga Lake, near Hendersonville, N. C., July 6, 1935 (L. C. Thomsen). No. 1463.1 in Cornell University Collection.

Allotype—Female imago. Florence, S. C., May 16, 1929 (O. L. Cartwright). No. 1463.2 in C. U. Collection.

Paratypes—Three female imagos, four male subimagos; same data as allotype. No. 1463.3-9 in C. U. Collection.

Genus *Ephemera* Linn.

Ephemera guttulata Pict.

Another species reported for the first time from this state. This handsome and strikingly-marked *Ephemera* was fairly abundant at Valle Crucis. Adults were taken just after dusk, flying low over the creeks, upstream and back again, rather after the fashion of small dragonflies hawking mosquitoes. Only a pale streak could be seen as one of them passed by. The dark-mottled wings contrast sharply with the white body, and are practically invisible in the twilight. Specimens of both sexes were obtained as they repeatedly passed forward and back above the stream bed; however, females were more numerous than males. Most of the specimens were taken May 25-27, although others were seen after that period. A nymph of this species was taken by Prof. Needham from Twenty-Mile Creek, Great Smoky National Park, on Apl. 3, 1934.

Ephemera blanda Trav.

More abundant than the preceding species, and evidently more widely distributed in the state. This species usually flies much higher than *E. guttulata*, but is taken at the same time, just after dusk. It is often found at some distance from any stream. Sometimes great numbers of *E. blanda* are seen flying above a highway, all going in the same direction, and most of them too high to be taken in a net. Returning to Valle Crucis from Heaton, one evening just after sunset, we stopped the car to find out what insects were coming up the road toward us in such numbers. By leaping as high as possible, we were able to catch a few of the low-flying ones, which proved to be specimens of *blanda*. All that we caught that evening were males. Another evening, from just

before sunset until it was too dark for collecting, males and females were taken as they 'danced' above and along the shores of Wildcat Lake, a small lake near Banners Elk. Collections are as follows. Imagos: Valle Crucis, May 25-27, 1936 (J. R. T.) and May 30, 1935 (L. C. Thomsen); Wildcat Lake near Banners Elk, May 31 (J. R. T., L. C. T.); Heaton, June 2; Banners Elk, June 8; Cedar Mountain, June 13 (J. R. T., L. C. T.). Nymphs: Valle Crucis, late autumn, 1934, and Oct. 1, 1935 (L. C. Thomsen); Hazel Creek, Cataloochee Creek, and Twenty-Mile Creek, Great Smoky National Park, Apl. 3-7, 1934 (J. G. Needham).

Subfamily Potamanthinae

Genus *Potamanthus* Pict.

Nymphs of this genus were found in the Davidson River, Pisgah National Forest, not far from the place where *Oreianthus purpureus* was first taken in 1929. *Oreianthus* nymphs inhabited the deeper, swifter portions of the river, beneath very large, flat rocks which could be moved but slightly and with much difficulty. *Potamanthus* nymphs, however, occurred in the shallower, less rapid waters nearer the bank. Although some dwelt in the fine gravel and sand in the lee of smaller rocks, others were clinging to the sides of the rocks, not loosening their hold when their support was lifted from the water. They seemed to prefer angular, sedimentary rocks in water not over one foot in depth, to which they clung, heads upstream, an inch or two above the stream bed.

Potamanthus distinctus Trav.

This species has not been listed hitherto from North Carolina. The nymph has not been described previously. I was able to rear a male and a female adult from nymphs, and thus to determine the species.

Nymph. Body of female, 17 mm.; of male, 14 mm. Head deep red-brown with pale markings. Mandibular tusks of the type of *P. rufus* Ide*, but with fewer spines on the distal portions (see fig. 7). Anterior to median ocellus, a large, pale, mushroom-shaped spot, and two small lateral streaks at its antero-lateral angles. Small pale spot behind median ocellus. Large pale areas around eyes. Median line of occiput pale; pale transverse streaks along this line; two pale sub-median spots on hind margin. Eyes separated by a space approxi-

* 1935—Ide, F. P. Life history notes on Ephoron, Potamanthus, Leptophlebia and Blasturus with descriptions (Ephemeroptera). Canad. Ent. 67: 121, figs. 6, 6a, Pl. 4.

mately equal to three eye diameters. Thorax deep red-brown. Sagittate median mark on prothorax; lateral margins widely pale; a pair of submedian triangular spots, two pairs of pale submedian streaks, and smaller pale marks laterally. Mesonotum with many pale marks, principal of which are: two pairs of submedian marks, anterior pair elongated, posterior pair rounded; two pale spots on wing roots; four or five other pale spots between wing roots and anterior margin.

Fore tarsus red-brown; wide blackish band at base, narrower dark band apically. Tibia yellowish; blackish median band, also narrow black streak on outer margin near base. Tibial spine quite short. Femur yellowish; wide blackish pre-apical band; apical flange dark; blackish band near base, on outer half only, connected along outer margin with pre-apical band. Long hairs on inner margins of femur and tibia; shorter spines on tarsus. Middle and hind legs yellowish. Base of tarsus dark brown; narrow dark band at base of tibia, dark pre-apical mark on outer margin; pre-apical and basal bands on femur, as in fore leg. Abdomen dark red-brown dorsally, yellowish ventrally. Pale dorsal markings of abdominal tergites 3-8 consist of a short, median dash at anterior margin, and two submedian triangular or conical spots on posterior margin, faint traces of 2 or 3 small dots near lateral margin. On tergites 9 and 10, submedian marks on anterior margin, in addition to the above. Sternites 3-8 each with a curved, brown, lateral mark; a pair of brown submedian spots near anterior margin; posterior to these, a pair of smaller dark dots nearer the median line. Gills pale purplish brown. Tails dark red-brown, paler distally.

The mature nymph of *P. distinctus* is quite similar to that of *P. rufus*. Slight differences in the mandibular tusks, in the coloration of legs and abdominal tergites, and the presence of more extensive dark ventral markings, serve to distinguish it from the latter species. Adults of both sexes, in *distinctus*, possess blackish cross-veins in the wings; in *rufus*, wing-veins of both sexes are wholly pale, and eyes of the male are somewhat smaller. In both species, lateral ruddy markings occur on the abdomen. This reddish coloration of the adults fades very quickly in alcohol, so that the female of *distinctus*, which is so preserved, has already lost the ruddy abdominal markings, the median red stripe on head and thorax, and the ruddy tinges of the fore legs, which were present at the time of emergence. The two male specimens taken last summer are pinned, to preserve the ruddy coloration. It is probable that the female specimens to which I referred in a previous paper* as *Potamanthus* sp. No. 2, are *P. distinctus*.

* 1932—Traver, J. R. Mayflies of North Carolina, Pts. I and II. J. Elisha Mitchell Sci. Soc. 47: 110.

Immature nymphs taken along with the mature ones just described, differ somewhat in the length of the tusks and in the maculation of the abdominal tergites. Color pattern of head is identical with that of the mature nymphs; leg pattern also similar, although the femora appear relatively stouter. Similar dark ventral markings are present. I am considering these nymphs to be of the species *distinctus*. An examination of many specimens of nymphs of *P. walkeri* Ide, from the Potomac River at Brunswick, Md., leads me to believe that the color pattern of the abdominal tergites may be quite variable within the limits of a single species. It would seem that care should be taken to indicate the sex as well as the relative maturity of a nymph, when describing the color pattern.

Subfamily Neophemerinae

Genus *Oreianthus* Trav.

Oreianthus purpureus Trav.

This unique species has been reported to date from three localities only, in North Carolina: the Mitchell River near Mountain Park, Surry Co.; the Tuckaseegee River at Dillsboro, Jackson Co.; and the Davidson River in Pisgah National Forest, Transylvania Co. This past summer I succeeded in rearing several male imagoes from the Davidson River, so that nymphs and imagoes of both sexes are now known. I note that, in previous descriptions of the nymph, no mention has been made of a pair of low, submedian tubercles on the anterior margin of the pronotum. The short, median spine or tubercle on the mesonotum, between the wing cases, was in error stated to be on the metanotum.

Male imago (alcoholic specimen). Body 14 mm ; wing 14-15 mm.; tails 20 mm. Eyes large, rounded; not contiguous apically; separated by about $1\frac{1}{2}$ mm. Lower portion very small, blackish, quite distinct from the large upper portion, which is orange in color. Head and antenna purplish black; the latter with a whitish ring at base. Pronotum deep purplish, lateral margins blackish; posterior margin quite deeply emarginate. Mesonotum deep red-brown; paler laterally, median stripe somewhat paler. A cream-colored line along antero-lateral and postero-lateral margins, on each side; another pale curved line on each side, extending from just above the antero-lateral margin to a point anterior to the scutellum, thence backward on each side toward scutellum. Scutellum and preceding area piceous. A very small tubercle on median line, about the middle of the scutum. Posterior notal

wing processes produced backward somewhat beyond the scutellum; tips somewhat pointed. Metanotum purplish brown. Thoracic pleura deep red-brown, with numerous large pale areas; purplish red shading above and anterior to each leg. A wide purplish black streak extends forward from base of fore wing; a narrower and shorter blackish streak just below it; another black area anterior to middle leg. Sternum deep red-brown; middle of prosternum blackish; intersegmental areas pale.

Fore leg not quite as long as body. Femur purplish red, paler basally, blackish apically; several small pale dots or streaks, on purplish median portion. Tibia piceous; tarsus somewhat paler than tibia, deep purplish brown. 'Knee' pale. Femur about $\frac{3}{4}$ as long as tibia, which is approximately $\frac{3}{4}$ the length of the tarsus. Basal fore tarsal joint very short; second and third joints subequal; fourth joint slightly shorter than third; fifth joint about $\frac{1}{2}$ as long as third. Middle and hind legs very similar to fore leg in coloration. Pale spots on femur more numerous; extreme base whitish. Knee area more conspicuously whitish. Tibia and tarsus concolorous, deep purplish brown; tip of tibia creamy. In hind leg, tarsus only a little more than $\frac{1}{2}$ as long as femur; tibia very slightly longer than femur, fully twice the length of the tarsus. Distal joint of hind tarsus about equal in length to basal and second joints combined. Third joint shortest, second slightly longer, basal slightly longer than second. Wing membrane very faintly milky in most specimens. Costal and subcostal spaces beyond bulla with an almost opaque white cloud, becoming paler at apex. In general, very similar to wings of female. Costal cross veins very weak, especially at base and in stigmatic area; in latter space, strongly anastomosed, dividing the area into two sets of cells, of which the outer series is slightly the smaller. All veins deep purplish black; longitudinal veins heavier than cross veins, costa, subcosta, and radius the heaviest. Humeral cross vein faintly purplish.

Tergites and sternites deep purplish red, with numerous pale dots. Antero-median portions of tergites 2 and 3, and antero-median margins of tergites 4-6, pale. A black median streak on tergites 1 and 2; on 3-6, narrow black submedian streaks enclosing a paler median line; on 7-9, *pale submedian* streaks enclose a *dark median* line. Tracheae faintly etched in paler color. Posterior margins of all segments, both tergites and sternites, pale greyish white, so that abdomen appears distinctly annulate. Intersegmental areas likewise pale. These pale margins widest on sternites 6-8. Pleural fold deep purplish black, except at posterior and anterior margins; blackish shading on each side, on tergites and sternites. A blackish transverse streak across tergite 2.

Middle area of forceps base purplish red; lateral portions paler. Basal portion of forceps, penes, and lateral margin of tergite 10, pale yellowish white, shaded with purplish grey. Apical portion of forceps purplish grey. Forceps *four-jointed*, not three-jointed, as seemed to be indicated in the male nymph. The two distal joints very short; basal joint about $\frac{1}{4}$ as long as the second, distinctly concave on the inner margin, and bearing a protuberance at the inner apical angle (see fig. 8). Penes much as in *Neophemera bicolor* McD. Tails very deep purplish black; in basal portion, a very narrow and inconspicuous paler ring at each joining. All three tails subequal in length.

Male imago (dried specimen). Eyes deep purplish red. Entire thorax deep mahogany brown, very shiny. Pale areas on pleura quite bright yellow; most conspicuous are three stripes (including anterior margin of mesonotum) anterior to wing roots, separated by wider red-black streaks. Tibia of fore leg, and all parts of middle and hind legs, concolorous with thorax; pale spots on femora inconspicuous. Coxa and trochanter of fore leg, and coxa of middle and hind legs, yellowish with purplish red shading. Base of fore femur yellowish. 'Knees' of all legs yellowish. Wings hyaline, no trace of milky tinge seen in alcoholic specimens. A distinct yellow area on membrane at base, on anterior and posterior margins of both wings. Abdomen deep purplish red; pale posterior margins of segments not conspicuous; pale spots barely noticeable.

Female imago (dried specimen). Very similar to male, but head and thorax distinctly dark purple with yellow mottling. Yellow areas on pleura, legs, and wing bases even larger and more conspicuous than in male. Pale spots on femora more in evidence. Posterior margins of all abdominal segments distinctly yellow, likewise anterior portion of pleural fold on segments 8 and 9, and entire pleural margin of 10. Tails deep purplish black.

Allotype—Male imago, reared from nymph. Davidson River, Pisgah National Forest, June 12, 1937. No 1002.2 in Cornell University Collection.

Nymphs of *Oreianthus* are quite hardy, able to withstand unfavorable conditions for a considerable time. Early in June I made a trip to Davidson River from Valle Crucis, at which time four nymphs of this species were captured. On the return trip the nymphs, in a rearing cage, were placed in a pail partly filled with water and carried thus in the car for several hours. Fresh water was provided only twice during the trip. All four nymphs survived the trip, likewise another trip from Valle Crucis to Penrose on June 10. Three of the nymphs died

on June 13. Another nymph survived a long, hot trip of three days (June 26-28) from Penrose to Tuscaloosa, Ala. During this time, the water was changed but a few times each day. The pail, however, was taken out of the car each night and set on the running board. This nymph died on June 29.

From June 11 to June 24, Dr. Lillian Thomsen and I collected seventeen *Oreianthus* nymphs from the Davidson River. Collections were made at three different points in the main stream, and in one small tributary. The nymphs were usually found beneath small, isolated boulders or irregularly-shaped sedimentary rocks in rapid water. In 1929 and 1930, I had found them only under the much larger, flat rocks which form the main bed of the stream. In collecting, one of us held the hand screen downstream from a given rock, whilst the other lifted the rock enough to permit the current to sweep under it. Sometimes two or three nymphs were found under one rock; again, half an hour's work yielded not a single specimen. In one instance only was it possible to determine the position of the nymph in reference to the sheltering rock. In the small tributary just mentioned, a good-sized rock was lifted from the water, and beneath it, clinging to the under surface in the manner of the large stonefly, *Pteronarcys*, was one *Oreianthus* nymph. It did not loosen its hold, even after the rock was well out of the water. Although the current was fairly swift here, the water was shallower than in most points in the main stream, probably not over nine inches in depth.

Oreianthus nymphs have a habit of 'playing dead' when disturbed, much after the manner of nymphs of the genus *Ephemerella*, so that it was not always possible at first glance to be certain that a bit of apparent trash on the hand screen was not a nymph. The same habit was noted in nymphs kept in the rearing cages.

Although we made frequent visits to the river during these days in June, at varying periods of time between 9:30 a.m. and 9 p.m., often watching for several hours at a stretch, we did not see a single *Oreianthus* adult in flight. Nor were any found resting on trees or shrubs along the bank of the river. As to the time of transformation from nymph to subimago, I record my observations on those nymphs kept in the rearing cages at Penrose. Some subimagos emerged in the forenoon, between 8 and 9:30 a.m. Others emerged in the evening, 8 to 9 p.m. One male emerged about 2:45 p.m., one female at 11 a.m. Is it characteristic of the species, that the individuals emerge at such varying times during the day? Is this lack of uniformity in time of

emergence due to the fact that the nymphs were in captivity, or do they behave similarly when undisturbed, in their native haunts?

At Penrose, we made our home in a cabin part way up the side of Fodderstack Mt. Above and behind the cabin, in a spot sheltered by young trees and shrubs from too direct sun-light, the nymphs were reared. The rearing cages were kept in a small tub under the continuous drip from a leaky pipe-line conducting water down from a spring near the top of the mountain. This proved an ideal situation for the rearing of nymphs. Subimagos, when removed from the rearing cages, were put into similar dry wire cages, each containing a small leafy branch of tree or shrub. Sourwood, being abundant, was used frequently for this purpose. Sufficient moisture for transformation of the subimagos was provided by the leaves on this small shoot. Another leafy branch was placed on top of the cage, which was then taken inside the cabin, for protection from sudden showers.

The following item from my field notes might be of interest. "Did not remove two nymph skins until the afternoon of the day on which the subimagos emerged early in the forenoon. Could then find no trace of either whole skin, only the abdomen of one, and small bits of thorax, presumably of the two skins. It looks as though the other nymphs (*Oreianthus* and two *Irons*, in the same cage) must have eaten them." Nymph skins of *Oreianthus* were usually recovered whole; further, the force of water in the tub was not sufficient to have caused the disintegration of the skins. Even then, portions of the abdomen of the second skin would have been found in the cage.

The nymph of another species of *Oreianthus* is described elsewhere in this paper, under Florida.

Family HEPTAGENIIDAE

Genus *Stenonema* Trav.

Stenonema carolina Bks.

Imagos were taken in flight at Cranberry, June 8 (L. C. Thomsen, J.R.T.); at Valle Crucis, May 26-June 9; at the Davidson River, June 16 and 20; and near Banners Elk, June 8. A few specimens were reared at Valle Crucis. Nymphs are from a stream between Boone and Blowing Rock, June 7; near Banners Elk, June 3; and Valle Crucis, May 30. A small, pale, male imago from Conestee Creek, near Cedar Mountain, probably belongs here.

The characteristic greenish cast of the bodies of the male imagos fades quickly in alcohol. There is considerable variation in size no-

ticeable among pinned as well as alcoholic specimens. Associated with the normal specimens,—bodies greenish yellow, posterior margins of both tergites and sternites blackish,—one finds smaller, paler forms,—bodies yellow, only the tergites darkened posteriorly. Markings of head, thorax and legs are similar to those of the normal dark specimens; eyes are as far apart, genitalia exhibit no differences. I am holding such forms as pale varieties of *carolina*.

The nuptial flight of the males was witnessed beside a small stream near Cranberry, on June 8. The highway passes close to the stream on one side; on the opposite bank grow many tall trees. The greenish-yellow bodies of the males, their wings reflecting the sunlight, were noticed some time before sundown, weaving up and down in the manner of most species of mayflies. Most specimens were flying high, fully twenty feet up in the air, like tiny streaks of light against the background of dark foliage. A few minutes of flight alternated with a resting period, when the insects perched on some leaf of a nearby tree, visible to us, but quite safe from our nets. Occasionally one dropped low enough to come within range of the net. Many were dancing high above the roadway; several of these were captured as they dropped down to rest on some low-growing shrubs near the road. Now and again a mating pair was seen; one or two were captured. Several females, flying low over the stream in oviposition, were also taken. Both sexes, however, were quick to elude the net of the would-be collector.

***Stenonema pallidum* Trav.**

A few imagos which seem to be of this species were reared from nymphs taken at Valle Crucis, May 27–June 7, and at Banners Elk, June 3. Two nymphs are from Valle Crucis, June 7. Imagos were taken in flight at the Davidson River, June 16–20 (L.C.T., J.R.T.). From these pinned specimens, I present the following additions to the original description.

Male imago (dried). Body 8 mm.; wing 9 mm. Eyes deep purplish red. Head and thorax deep yellow, the latter almost orange on mesonotum, metanotum, and portions of pleura. Femora deep, bright yellow; tibiae and tarsi duller and much paler. Median band on hind femur may be almost obsolete. Wings highly iridescent; costal and subcostal spaces distinctly amber-tinged. Dorsum of abdomen quite bright canary-yellow, posterior margins of tergites purplish black except near lateral margins. Apical tergites do not differ in color from those pre-

ceding. Venter paler and duller yellow, without dark markings. Tails very faintly ruddy at joinings, in basal half. Other markings as indicated in original description. Very short dark dash below antenna; lateral black streak on pronotum; no dark marks on pleura; no dark stigmatic marks. Basal joint of fore tarsus fully $\frac{1}{2}$ as long as the second joint.

Female imago (dried). Very similar to male. Thoracic notum and pleura not quite so deep in color. Median band of hind femur obsolescent. Costal margin of fore wing even more deeply amber-tinged; cross veins in basal costal space may be somewhat more heavily margined, and more numerous (5 or 6 in female, before bulla,—usually only 4 in male). Less contrast between dorsum and venter of abdomen, both being light canary yellow. Tails whitish, unmarked.

This species flies in the evening, at about the same time as *S. carolina*, and may be associated with it.

***Stenonema ithaca* Clem. and Leon.**

Imagos were taken at Davidson River, June 5–15. Nymphs are from the North Toe River near the little town of Minneapolis, June 8; Cove Creek near Sugar Grove (north of Vilas), June 9; stream near Blowing Rock, June 7. This species also flies at sundown; ovipositing females may be taken as late as it is possible to see them in their flight over the water. Males usually dance at a height of about 8 to 12 feet.

***Stenonema pudicum* Hag.**

As on previous collecting trips in the mountain region of this state, this species was the one most frequently met with. Imagos were taken as follows: stream near Blowing Rock, June 7; near Banners Elk, May 31–June 8; Valle Crucis, May 28–June 9; Cedar Creek near Glenville, June 19; Heaton, June 2. Reared specimens are from Valle Crucis, June 6; Cathey Creek at Cherryfield, June 21. Nymphs are from Valle Crucis and Foscue. Other nymphs were taken by Prof. Needham in the Great Smoky National Park at Hazel Creek, Cataloochee Creek, Twenty-Mile Creek, and Big Creek at Walnut Flats, Apr. 3–7, 1934.

This large, handsome, alert species with the beautifully marked wings was one of the principal actors in the brilliant performance staged at Valle Crucis on pleasant evenings in late May and early June, over Crabapple Creek. Other actors in this dance-drama were *Isonychia sadleri*, *Ephemera guttulata*, *Ephemera blanda*, *Iron rubidus*, *Rhithro-*

gena rubicunda, and *Rhithrogena amica*. These twilight flights of hundreds of mayflies commenced just as the sun was setting, and continued until darkness descended. Similar flights were witnessed during the same period near Banners Elk, where the highway crosses a small stream. Above the bridge the air literally teemed with the dancing figures of mayfly multitudes. Smaller species, of the Ephemerellas and Paraleptophlebias, also *Iron confusus*, often joined the throng. But of them all, *Stenonema pudicum* and *Isonychia sadleri* were the most elusive, the most difficult to capture.

Genus *Heptagenia* Walsh

Heptagenia junio McD.

This species has not been recorded previously from North Carolina. Two male imagos, several female imagos, and nymphs were collected. One male and two females were reared from nymphs. Localities: Valle Crucis, May 29–June 10 (adults and nymphs); Banners Elk, June 3 (adult females, nymphs); Davidson River, June 21 (adult male, nymphs); Cathey Creek at Cherryfield, June 17 (nymphs). Specimens were taken also in the Great Smoky National Park near Elkmont, Tenn.

Heptagenia aphrodite McD.

Specimens are from Valle Crucis, May 27–29 (adult females); Cove Creek near Sugar Grove, June 9 (nymphs); and a tributary of the Rocky Broad River at Lecky Gap, June 16 (nymph). See also a doubtful record of this species from Alabama.

Heptagenia thetis Trav.

Nymphs were collected at Banners Elk, May 31, and at Heaton, June 3. Other specimens were taken by Prof. Needham in Twenty-Mile Creek, Great Smoky National Park, Apl. 3, 1934.

Heptagenia marginalis Bks.

Nymphs were taken in a stream near Blowing Rock, June 7.

Heptagenia julia Trav.

Male and female imagos were reared from nymphs taken in Lecky Gap, June 19–21. Nymphs were also taken near Blowing Rock, June 7. Three of the imagos were pinned, and from these dried specimens it is possible to add the following notes to the original description of the species.

Male imago (dried). Vertex and occiput of head deep red. Prono-

tum dark red-brown in median area, lateral areas yellow. Mesonotum red-brown, tip of scutellum blackish. Pleura paler red-brown in a diagonal band from base of fore leg to base of hind leg; remaining areas yellowish. Prominent black markings above and posterior to fore leg, anterior to and above middle and hind legs. Sternum yellow. Fore femur yellow, tinged with red-brown at base; wide median red-brown band; black line along inner margin apically. Tibia and tarsus light olive brown, tibia with reddish tinge; tip of tibia blackish. Femora of middle and hind legs yellow, no red tinge; median band inconspicuous on hind femur; on each, a short, black dash near apex, on inner margin. Tibia and tarsus yellowish olive. Wings with a faint yellowish or amber tinge. A wide, red-brown band occupies the middle of the abdomen dorsally. Wide, black, posterior margins on tergites in this area. Lateral areas of tergites paler red-brown. Sternites deep yellow; posterior margins purplish red. Segments 8 to 10 with distinct reddish tinge. Tails olive yellow, joinings black.

Female imago (dried). Very similar to male. Tibiae and tarsi of all legs pale yellowish olive. Wings more distinctly amber-tinged than those of male; along the costal margin they appear greenish yellow. Dorsum of abdomen somewhat paler than in male, purplish red; pleural fold pale, likewise lateral margins of tergites 7-10.

Heptagenia spinosa Trav.

Two female imagos, taken at the same time and place as the type male specimen, were found in a vial with specimens of *H. aphrodite*. There seems no reason to doubt that these are the females of *H. spinosa*, hitherto undescribed.

Female imago (alcoholic specimen). Body 7 mm.; wing 7 mm. Yellowish. A purplish black spot between eye and lateral ocellus. Blackish mottling on occiput. Antennal filament somewhat dusky. Median portion of posterior margin of pronotum blackish; blackish markings on posterior half of pronotum, along median line. Mesonotum deeper yellow; scutellum pale. Posterior and lateral margins of metanotum, and triangular areas on each side of pale median line, purplish black. Purplish shading at bases of wings. A narrow, blackish line at apex of fore femur, another along inner margin near apex. Tarsal joinings and line at apex of tibia, narrowly black; distal joint of tarsus, and claws, dusky brownish. Other legs similar, but dark lines at joinings less conspicuous. Basal joint of hind tarsus slightly longer than the second joint. Longitudinal veins in basal half of fore wing pale yel-

lowish, except in cubito-anal region, where they are silvery white; cross veins not visible. In apical half of wing, both longitudinal and cross veins are visible, pale yellowish brown; longitudinals slightly heavier than others.

A wide, dark purplish band occupies the posterior half of tergite 1. Tergite 2 largely overlaid with purplish grey shading, leaving as pale areas the lateral margins, median line, and submedian semicircular spots on anterior margin. Median area of posterior margins of tergites 3-6 purplish. Purplish brown shading on median portion of subanal plate. This plate is somewhat conical, margin entire; extends slightly beyond the apex of tergite 10. Tails yellowish, basal half tinged with brown; joinings in this portion purplish black.

Allotype—Female imago. North Fork, Swannanoa River near Black Mt., N. C., June 30, 1930. No. 1115.2 in C.U. Collection.

The type locality was re-visited during 1936, in the hope of obtaining nymphs and more imagos of this unusual species of *Heptagenia*. I quote from my field notes regarding the present condition of this stream, which is the type locality also for several unique species of aquatic insects described by Dr. Banks. "Fished in Swannanoa River at Black Mt. (no luck, very little aquatic life), and in the North Fork. The latter is entirely ruined, in the part where I used to fish, apparently by silt pollution. A gravel company is excavating a big area beside and above it; the course of the stream has been changed, and the aspect of the whole is different. Found practically no aquatic life but snails; the stones are covered with a dirty-looking sediment; the stream does not look clear and inviting any more." It is most unfortunate that this beautiful stream, once so rich in aquatic life, has become so barren.

Genus *Rhithrogena* Eaton

In 1929 and 1930, I obtained partial life histories of four species of this genus, all of which seem to belong to the *anomala* group. These four were described as new species, while two others, of which only the nymphs were known, were designated respectively as *Rhithrogena* sp. No. 1 and *Rhithrogena* sp. No. 2. One of the first group, *R. uhari*, is a piedmont species, and probably does not occur in the Appalachian region. In 1936, four species and a possible fifth were taken in the mountain area of the state. Two have not been recorded before from North Carolina; one of these is a new species. Members of the genus *Rhithrogena* seem to be quite local in distribution, as no individuals of the species *R. exilis* or *R. fuscifrons* were taken during this season's

collecting. The type localities were not visited, as headquarters were too far from these points to make feasible the transportation of nymphs. Both *Rhithrogena* and Iron nymphs die quickly when transported far from their native streams, hence I was unsuccessful in completing these life histories.

***Rhithrogena amica* Trav.**

This species, described from specimens taken near Ithaca, N. Y., is represented in material collected in the vicinity of Valle Crucis. It is a new record for the state of North Carolina. Careful comparison of nymphs and imagos, the latter both dried and in alcohol, with the type material, shows that there is little difference between the New York and the North Carolina forms, aside from the smaller size of the latter specimens. The imagos from North Carolina are somewhat more ruddy as to leg and body markings, but are otherwise similar. A few specimens were reared, but in several cases it was not possible to find the nymph sloughs in the rearing cages. In *Rhithrogena*, these sloughs are either very fragile, so that they disintegrate quickly in flowing water, or else they are fed upon by other nymphs in the cages. Imagos were captured in flight, and many nymphs were taken. Imagos: Valle Crucis, May 30–June 10 (L. C. Thomsen, J.R.T.); Heaton, June 3; near Banners Elk, June 8; Cranberry, June 8. Nymphs: Valle Crucis, May 25–30; Heaton, June 3–6; Banners Elk, May 31. Other nymphs were collected by Prof. Needham on Apl. 3, 1934, in Twenty-Mile Creek, Great Smoky National Park.

***Rhithrogena fasciata* Trav.**

Mature nymphs of this species were quite numerous in Cove Creek, north of Vilas, on June 9. From these, one male and four female imagos were reared. The female of this species has not been described previously. A single nymph, taken near Blowing Rock, on June 7, is probably of this species.

Female imago (alcoholic). Body 8–8½ mm.; wing 8–9 mm. Head flesh-colored. Usual small dark mark below antenna; one large and one or two small black dots on each side, beneath clypeus. Narrow, median, purplish line on median carina (not present on one specimen). A faint dusky or pale purplish band across back of head between eyes, extending forward laterally along margin of eye. Antennal filament dusky in basal portion. Pronotum flesh-colored; anterior, posterior, and lateral margins narrowly darkened; a short, dark streak in postero-lateral angle. In well-marked specimens, a dusky triangular area on

each side of median line. Mesonotum greenish yellow; scutellum and two small areas anterior to it brownish, in dark specimens; postero-lateral fold, and narrow line along antero-lateral margin, dull purplish. Metanotum greenish yellow laterally, median portion brown. Pleura greenish yellow; a dull purplish streak anterior to wing base; brownish markings above legs. Sternum flesh-colored; posterior half of mesosternum, and outlines of other median sclerites, red-brown.

Legs greenish yellow. Customary dark spot on each femur; short, black, longitudinal streak near apex, on inner margin; narrow dark band at apex. Apex of tibia, outer portion of tarsal joinings, apex of distal tarsal joint, and claws, dusky. Wings hyaline. Longitudinal veins in both wings light purplish brown, paler in anal areas; humeral cross vein somewhat deeper in color. Cross veins along costal margin and in apical portion of fore wing pale brownish, elsewhere almost invisible.

Dorsum of abdomen red-brown with a distinct purplish tinge. Inter-segmental areas and lateral fold, whitish. Pale median line on each tergite; pale, oblique, submedian streaks on basal and middle tergites, and a pale dot near end of each; on apical tergites, pale submedian dot only. Posterior margins of tergites darkened in middle area, paler laterally. On each tergite, near lateral fold, an irregular, scroll-shaped, dark mark. Sternites much paler than tergites; pinkish, with no dark markings except an indistinct brownish line on basal segments, near lateral fold. Posterior margins narrowly pale whitish. Tails yellowish, joinings narrowly purplish red.

Allotype—Female imago, reared from nymph. Cove Creek, north of Vilas, N. C., June 9, 1936. No. 1121.2 in Cornell University Collection.

The male imago from Cove Creek is somewhat larger and stouter than the holotype,—wing 8 mm. instead of 7 mm. (incorrectly given in original description as 6 mm.). The following additional notes on the species are presented from this more recent specimen. Thorax and apical abdominal segments red-brown, contrasted with the purplish brown basal and middle segments (type specimen paler, less contrast in color). Laterally on each tergite, a semicircular paler area is enclosed by a darker line, which is widened on the inner margin into an oblique streak extending inward and backward from the antero-lateral angle. Pale submedian streaks and dots on tergites, as in female. Sternites very pale purplish brown, apical ones darker; brown lateral streaks on basal segments; ganglionic areas faintly smoky. Tails pale, but tinged

faintly with red-brown at base; joinings purplish. A re-examination of the holotype shows that the tails, instead of being wholly whitish and unmarked, as stated in the original description, are brown-tinged basally, joinings very faintly darker. Longitudinal veins of the holotype are faintly tinged with purplish, not wholly yellowish, as originally stated; all cross veins invisible. I present a new figure of the genitalia of the holotype, drawn after these structures were treated in KOH and re-mounted (see fig. 4).

***Rhithrogena rubicunda* sp. nov.**

A small reddish species of the *anomala* group, allied to *R. fasciata*.

Male imago (specimen in alcohol). Body 8 mm.; wing 8-8½ mm. Head yellowish, with ruddy shading between eyes on posterior margin; a faint purplish line along median carina. Black semicircular mark below antenna; one large and one small black dot beneath clypeus. Antenna dusky; apex of basal segment black-ringed. Thorax reddish brown, notum and sternum darker than pleura. Pronotum margined and shaded with purplish black. Scutellum, two large areas anterior to it, and posterior margin of mesonotum on each side of scutellum, deeper brown. Anterior to wing roots, two purplish streaks: one directed obliquely laterad, the other extending along antero-lateral margin of mesonotum. Metanotum blackish brown except for paler anterior and lateral margins. Pleura light red-brown; a few dark brown markings above leg bases. Posterior margin of prosternum purplish black; middle sclerites of other segments of sternum outlined in dark brown.

Legs yellowish amber. Fore tibia with faint ruddy tinge. Narrow band at apex of fore femur, tip of tibia, joinings of basal and second tarsal joints, claws, and most of distal joint of tarsus, rather dark red-brown. Usual purplish black median spot on femur. On middle and hind legs, the tarsi are faintly shaded with ruddy brown; all tarsal joinings dark red-brown. A small, black mark near apex of each femur; no apical band. Median spot on hind femur somewhat elongated. Wings hyaline. Longitudinal veins and humeral cross vein light purplish brown, those of costal margin heaviest. Cross veins in anterior and apical areas of fore wing similarly colored but fainter, not as heavy as longitudinals but plainly visible. Cross veins elsewhere in both wings paler, becoming silvery white and almost invisible in anal areas. About 4 costal cross veins before the bulla, each sagged basad in middle; beyond bulla, about 3 cross veins before stigma. 14 or 15

stigmatic cross veins, more or less anastomosed, several of them somewhat aslant; faint milky cloud in stigmatic area.

Abdomen dark purplish red dorsally; paler ventrally, but with distinct purplish red tinge; semi-hyaline. Apical segments often with faint yellowish shading. Posterior margins of all tergites reddish black, of all sternites deep rose to purplish red, so that abdomen appears annulate with reddish. Pale, somewhat semicircular areas near lateral margin on each tergite, enclosed by darker line, are much less prominent than in *R. fasciata*. A pair of submedian, pale dots on each basal and middle tergite; on basal ones, traces of a pale median line in anterior portion. Lateral fold brownish; sternites pale next to this fold; ganglionic areas faintly indicated as whitish spots. Tails yellow, somewhat ruddy at extreme base; all joinings narrowly deep purplish red. Genitalia as in fig. 1.

Female imago (specimen in alcohol). Body 8 mm.; wing 9-9½ mm. Similar to male, except as indicated. Head often without ruddy or dusky shading (see notes on paratypes, however, particularly pinned specimens). Thorax somewhat paler than in male, yellow-brown with an olive tinge. All legs similar to middle and hind legs of male, but tarsi not noticeably darker than other joints. Longitudinal veins with a distinct amber tinge; cross veins visible also in disc of wing. Abdomen more or less opaque, except in spent females. Venter less distinctly ruddy-tinged, but reddish annulations of all segments evident. Egg valve and 8th sternite purple-tinged. Tails usually not ruddy at base. Lateral margin of 9th tergite with more or less purplish red shading. Considerable variation, also, as to depth of color of both thorax and abdomen; some paratypes are fully as ruddy on tergites and sternites as the type male. Others show ruddy or powder-white markings on pleura. In one specimen, the scutellum and adjacent areas are ruddy; distinct ruddy shading also, on posterior part of head.

Male (dried). Entire head, except narrow frontal margin, flushed with red. All femora very deep amber; fore tibia without reddish tinge; tarsi duller and paler than tibiae. Wings highly iridescent. Veins appear much paler than in alcoholic specimens. Dorsum of abdomen dark red-brown with purplish tinge; apical tergites bright red-brown. Greater contrast in color between dorsum and venter than in alcoholic specimens. Pale dots and pale lateral areas on tergites almost invisible. Anterior margins of basal and middle tergites narrowly pale; sternites 8 and 9 brighter reddish than preceding ones. In one specimen, from Valle Crucis, eyes deep purplish black (wholly

blackish in others). Scutellum and adjacent areas distinctly reddish. Tergite 10, and lateral margins of 9, bright yellowish orange. Wing veins almost whitish, except those on costal margin.

Female (dried). Entire head reddish. Distinct contrast between dorsum and venter of abdomen. Tergites 1-6 dull, dark red-brown with purplish tinge; 7 slightly paler; 8-10 much paler, with yellowish instead of purplish tinge. Venter paler purplish red.

Holotype—Male imago (dried). Banners Elk, N. C., June 5, 1936 (J. R. Traver). No. 1458.1 in Cornell University Collection.

Allotype—Female imago (dried). Banners Elk, N. C., June 8, 1936 (J. R. T.). No. 1458.2 in C. U. Collection.

Paratypes—Seven male imagos, eleven female imagos; Valle Crucis, N. C., May 25-June 8, Banners Elk, N. C. June 5-8, 1936 (J. R. T., L. C. T.). Some dried, others in alcohol. No. 1458.3-20 in C. U. Collection.

This species is about the size of *R. fasciata*, and of small specimens of *R. amica*. From the former it may be distinguished by the reddish annulations of the abdomen; less prominent lateral pale areas on tergites; cross veins on wing of male visible in anterior and apical areas. The distinct contrast in color between dorsum and venter of abdomen, and the almost total lack of reddish shading on pleura and femora, as well as the paler and less prominent wing veins, separate it from small specimen of *amica*. In genitalic type, it is quite close to *fasciata*; however, the apical portions of the penes are thicker, more rounded, while the spatulate process on each division of the penes is distinctly wider.

I do not know the nymph of this species, as imagos were taken only in flight. There is a possibility that the nymphs I am describing as *Rhithrogena* sp. No. 3 may be the immature forms of *rubicunda*. These nymphs occurred in considerable numbers in the same streams above which I took the adults of *rubicunda*. Seeming evidence against this theory is that the nymphs were not yet mature on June 10, although imagos of *rubicunda* were taken as early as May 25. The only mature nymphs taken from streams in this area, during the period of flight of *rubicunda*, were those of *amica*, and smaller specimens which I cannot distinguish from typical *amica*. These I assume to be the nymphs from which came the small specimens of apparent *amica*, mentioned previously. So similar in general appearance are the imagos of many species of *Rhithrogena*, that it was only after mounting the genitalia and comparing the specimen point by point in the laboratory that I realized there were two species among the specimens, all of which I had assumed to be *amica*.

Rhithrogena sp. No. 2

The nymph of this species was described in my previous paper on the Heptageniidae of the state.* A few of the orange nymphs were collected during 1936 from that tributary of the Rocky Broad River which flows through Lecky Gap; from these a single female imago was reared. A male subimago died when half out of its nymphal skin; genitalia not sufficiently developed to show details of structure. The femoral markings of the imago indicate that it is a member of the *anomala* group. Since the nymph slough of the female imago could not be found, and since further, the female bears a marked resemblance to those of two other species of this group, I am holding the specimens under the original designation.

Female imago. Body 7 mm.; wing $7\frac{1}{2}$ mm. Head pale yellowish; usual dark mark below antenna. A narrow purplish black line on posterior margin of head, slightly widened at each end, near eye. Filament of antenna dusky. Thorax, including scutellum, yellowish. Anterior and posterior margins of pronotum very narrowly darkened; a small area of purplish shading laterally, just above leg base. Posterolateral areas of mesonotum below scutellum shaded with purplish. Metanotum largely purplish brown. Posterior half of mesosternum brownish. On pleura, a small dark spot just above bases of middle and hind legs. Very minute dark dot at antero-basal angle of middle and hind coxae; on middle coxa, small dark dots also at postero-basal and postero-apical angles.

Legs pale yellowish; each femur with usual purplish black median mark (not streak). Tarsal joinings, claws, narrow line at apex of tibia, and somewhat wider mark at apex of femur, darkened. Wings hyaline. The first four longitudinal veins of the costal margin are pale purplish brown, as are also the cross veins between them; subcosta of hind wing similarly colored. All other veins and cross veins silvery white, the latter almost invisible.

Abdomen yellowish with pale pinkish tinge. Tergites 1-8 shaded with purplish, most apparent on tergite 3. Intersegmental areas paler; posterior margins of apical tergites appear narrowly darker. Lateral margin pale. Sternites paler than tergites; intersegmental areas whitish. Tails yellowish, tinged in basal portion with very pale red-brown; joinings narrowly darker.

* 1933—Traver, J. R. Mayflies of North Carolina, Pt. III. The Heptageniinae. J. Elisha Mitchell Sci. Soc. 48: 172.

The female imago is very similar to that of the allied species *R. exilis* and *R. fuscifrons*. From each of these it may be distinguished by the absence of definite darker posterior margins on the basal and middle tergites. The dark posterior margin of the head separates it from *exilis*; the entire body, particularly head and thorax, are much paler than in *fuscifrons*.

I note that in nymphs taken in the past season the entire head is thickly freckled with small dark dots; however, specimens from previous collections do not show this character. Posterior margins of all tergites narrowly darker. Two small dark, submedian dots near anterior margin; between and connecting them, a narrow, dark, transverse band. The nymph of *R. uhari* is likewise uniformly orange on the dorsum, but considerably paler than *Rhithrogena* sp. No. 2. The femora also are much paler, the head somewhat narrower, the gills (in life) distinctly grey. It is difficult to distinguish the nymphs of *Rhithrogena* sp. No. 2 from dark forms of *fasciata*, in alcoholic material, except for the deeper and more uniform orange color of the dorsum. I believe, however, that *Rhithrogena* sp. No. 2 is worthy of specific rank.

Rhithrogena sp. No. 3

Known only in the nymphal stage. A species of the *anomala* group; very similar to *R. fuscifrons* in general appearance, but more strikingly banded with alternating dark and pale areas.

Nymph. Body of immature nymphs, 6–8 mm.; tails 7 mm. Head as in well-marked specimens of *fuscifrons*. Large chestnut brown patch occupies middle area of head, from frontal margin backward between bases of antennae, and lateral ocelli, to posterior margin. Lateral portions yellowish, except for a light brown spot near outer margin, laterad of each eye. Antennae yellowish brown; in distal portion paler, joints shaded with purplish black, a black spot at each joining. Pronotum, and anterior portion of mesonotum to base of middle leg, dark red-brown; a few very faint paler markings on pronotum, two dark streaks on mesonotum between wing bases. Posterior portion of mesonotum, wing cases except at base, and entire metanotum, yellow. Fore and middle femora yellow in apical half or third; basal portion dark red-brown, with usual irregular pale median patch and purplish median spot. Tibiae yellowish, brown along outer margin, 'knees' brown; tarsi red-brown. Hind femur brown except for pale median streak in basal half; usual purplish median spot. Tibiae and tarsi as in other legs. Ventrally, dark brown markings near bases of fore and middle legs.

Abdominal tergites 1-2 and 8-10, wholly pale yellowish; tergite 7 may be wholly pale, or dark in basal half. Tergites 3-6, and sometimes base of 7, dark red-brown. Inner half of each gill on segments 2-5, and basal inner portion of gill on segment 6, deep purplish; gills of first and seventh pairs, and remaining portions of others, pale creamy white. Gill tufts purplish grey. In a female nymph slough, only the basal portions of gills on 2-5 are darkened. All sternites pale yellowish, unmarked. Tails yellowish brown; a narrow dark ring at each joining.

These strikingly-marked nymphs were quite numerous in the streams at Valle Crucis and in several streams near Banners Elk, from May 25 to June 10; up to this time, no mature nymphs had been seen. On June 11, I moved to Penrose. No nymphs of this species were found other than in the Valle Crucis region. A single female nymph slough was found in a rearing cage with a female imago, on June 6, and I supposed that this nymph slough belonged to the imago. Examination of it in the laboratory, however, showed that the wing cases were not long enough for those of a mature nymph. The female imago taken with it appears to be of the species *amica*, and I have no adults of *Rhithrogena* sp. No. 3. See note under *R. rubicunda*.

Nymphs of *Rhithrogena* sp. No. 3 bear a close superficial resemblance to those of *fuscifrons*. The following differences may be noted: (1) larger size; (2) pale strip including posterior half of mesonotum, all of metanotum, and the two basal segments of abdomen (in *fuscifrons*, entire thorax brown, usually also tergites 1 and 2); (3) brown apical area of third femur, as contrasted with yellow in corresponding areas of fore and middle legs (apices of all femora yellow in *fuscifrons*); and (4) the prominent purplish areas on the middle gills.

***Rhithrogena* sp.**

Adult females of an undetermined species of this genus were taken at Valle Crucis by Dr. Lillian Thomsen, May 27-30, 1935. These females are very similar to *R. amica* in general appearance, but larger. Body 10-11 mm.; wing 12-13 mm.

A single nearly mature nymph of a small species was collected in the Davidson River on June 20. Wholly whitish except for a very small median purplish mark on each femur, and a row of relatively long, dark spines on the posterior margin of each. Body 5 mm.

Genus *Cinygmula* McD.

***Cinygmula atlantica* McD.**

A new state record, unless this species is really synonymous with *C. subaequalis* Bks. Dr. Banks' specimens were taken from the North

Fork of the Swannanoa River in May. As stated previously, this stream supports no aquatic insect life at the present time. Mature nymphs of *C. atlantica* were found at Valle Crucis, Banners Elk, and Heaton, during the last week of May, but were by no means numerous. Attempts to secure a male imago were unsuccessful, but three females were obtained by rearing the nymphs, and one was taken in flight. The latter specimen is slightly larger than the reared forms, but similar in other respects. Females and nymphs have been compared with reared specimens of *atlantica* from the vicinity of Ithaca, N. Y. Imagos: Valle Crucis, May 28-30.

Genus *Iron* Eaton

In a previous paper* I have designated two groups of species in this genus, which occur in North Carolina, distinctions being based principally on nymphal structures. These are respectively, Group I, the *humeralis-rubidus* allies; and Group II, species of the *dispar* type. A third division, Group III, which includes species of the *longimanus* type, is represented in my collections of last summer. Nymphs of Group III may be characterized thus: (1) Gills of first pair have the anterior lobe greatly developed; this pair of gills is considerably larger than any succeeding pair; and the anterior lobes lie under the body of the nymph, tending to approach one another. (2) Gills of seventh pair tend to meet beneath body of nymph. (3) Head definitely widest near frontal margin, as in Group II. (4) Postero-lateral spines on middle abdominal segments relatively short and rather blunt, as in Group II. (These spines are present on segments 1-7, in all *Iron* nymphs, but are poorly developed on the basal segments.)

Species of the genus known to occur in North Carolina are as follows. Group I,—*I. rubidus* Trav., *I. subpallidus* sp. nov. (designated originally as *Iron* sp. No. 2), and *Iron* sp. No. 1, a piedmont form; Group II,—*I. dispar* Trav. and *Iron* sp. No. 4; Group III,—*I. confusus* Trav. and *Iron* sp. No. 5.

Iron rubidus Trav.

As originally described, males of this species ranged in body size from 8 to 9 mm.; females from 9½ to 11 mm. It should be noted here that the other specimens among the type material are somewhat larger. Thus, several males have a body and wing length of 10 mm., while some

* 1933—Traver, J. R. Mayflies of North Carolina, Pt. III. The Heptageninae. J. Elisha Mitchell Sci. Soc. 48: 156.

of the females reach 12 mm. in length. There is considerable variation also as to body markings and depths of color of the thoracic notum in the male. During 1936, I collected several nymphs of *rubidus* from Lecky Gap, others from Cherryfield, and from these was able to rear a few imagos. From a pinned male specimen (Lecky Gap, June 20) I present the following additions to the original description, which was drawn from a specimen in alcohol.

Male imago (dried). Eyes deep red-black. Entire head suffused with bright red. Thorax deep yellow. Pronotum largely suffused with bright red. Posterior and lateral margins and oblique lateral line, black. Scutellum and posterior margin of mesonotum blackish, as are the median projection and posterior margin of metanotum. Reddish shading anterior to wing base and on pleura above each leg. Black markings above leg base quite prominent. Narrow reddish line along anterior margin of mesosternum; median area of this sclerite red-tinged in posterior half. Fore femur largely ruddy; black bands prominent. Fore tibia with reddish flush; tarsus wholly pale yellowish white. On middle and hind legs, femur amber yellow, reddish only at apex; purplish streak on margin near median dark mark. Tibia red-tinged except in median area; tarsus pale. Abdomen yellow, venter rather paler than dorsum. Basal tergites shaded laterally with purplish. Middle and apical tergites with distinct rose-red lateral patches based on posterior margin, these being most prominent on tergite 8. Whitish area next to pleural fold, on tergites 9 and 10, and on lateral portions of sternites 8 and 9.

Many nymphs were taken in and near Valle Crucis, during 1936, which very much resemble those of *rubidus*, but seem a trifle larger; gills purplish red; color of body olive brown in life. From these, many imagos were reared, and various others of both sexes were captured in flight. Although showing some variation, both as to size and markings, from type material of *rubidus*, I can point to no character or constant marking by which either nymphs or imagoes may be distinguished from *rubidus*. The nymphs are doubtless those to which I referred in a previous paper as *Iron* sp. No. 3. I propose to regard these specimens, for the present at least, as *Iron rubidus*, dark form. Since, however, they may represent a distinct species, I present descriptions of both sexes, from pinned specimens.

***Iron rubidus*, dark form**

Male imago. Body $10\frac{1}{2}$ mm.; wing $10\frac{1}{2}$ mm. Head creamy white between eyes, elsewhere suffused with red. Antenna wholly dusky.

Thorax rather dark red-brown. Purplish red shading laterally on pronotum. Very deep red-brown median strip extends length of mesonotum. Scutellum, postero-lateral margins, and posterior margin, black. Anterior to scutellum, two paler reddish streaks. Prominent black markings on pleura; black streak above fore leg. Median portion of posterior half of mesosternum deeper brown, enclosing two reddish submedian streaks. Fore femur deep amber brown, faintly red-tinged; tibia paler, red-ringed; tarsus pale grey, last two joints dusky, all joinings and claws darker. Other legs similar but paler; no red flush on tibiae; apical half of tarsi distinctly dusky; usual dark mark on femora. Longitudinal veins yellowish; cross veins paler, but visible throughout most of wing.

Abdomen pale yellowish white, middle segments largely hyaline. Posterior margins of tergites narrowly purplish black; more or less pale brown shading laterally and along posterior margin (leaving area next to pleural fold whitish); small brown mark at each spiracle. Short black streaks on posterior half of median line, on middle and apical tergites; may be absent from basal ones. Tergites 8-10, and sternites 8 and 9, opaque, with alabaster markings on an orange-red background. Tips of forceps smoky. Tails smoky brown at base, becoming paler distally; indistinctly pale at joinings.

Variations of the above. All femora flushed with reddish brown; tarsi wholly dusky. Dorsum of abdomen more or less suffused, except in pale area next to pleural fold, with orange-brown, more intense on apical tergites. Rose markings on apical portion of forceps base.

Female imago. Body 10½-11 mm.; wing 12-13 mm. Head, posterior to lateral ocelli, creamy with very faint pink flush; anterior portion of head reddish. Pronotum reddish brown, margined and shaded with black. Remainder of thorax rosy, with extensive creamy white markings. Black pleural markings as in male. Fore femur red-brown, others amber-brown; usual dark median spot. Tibiae greyish, tinged at each end with lavender. Tarsi dusky at tips, elsewhere greyish, all joinings darker. Humeral cross vein almost wholly purplish (in male, pale next to costal margin). Dorsum of abdomen, except pale lateral area, flushed with brown or orange-brown (reddish, before eggs are laid) Otherwise as in male.

***Iron subpallidus* sp. nov.**

Iron sp. No. 2 in Traver, J. Elisha Mitchell Sci. Soc. 48: 160. 1933.

A member of the *humeralis-rubidus* group. Distinct by reason of

the relatively short postero-lateral spines on abdomen of nymph. Imago rather pale.

Male imago (alcoholic specimen). Body 11 mm.; wing $11\frac{1}{2}$ mm. Very similar to *I. rubidus*, but paler and somewhat larger. Head pale yellowish; bases of ocelli blackish; antennal filament dusky, two very small black dots beneath clypeus. Eyes slightly larger than in *rubidus*, contiguous apically (in some specimens of *rubidus*, eyes do not quite meet dorsally). Thorax pale yellowish brown, paler than in *rubidus*. Posterior and lateral margins of pronotum *not* darkened; lateral oblique dark dash on this sclerite reduced to a faint trace; very pale brownish shading on posterior half. Two purplish spots on pleura, above leg base. Scutellum of mesonotum, and posterior margin of sclerite on each side of scutellum, dark brown. A pale band along median area of mesonotum. Metanotum yellowish; median projection and posterior margin brown. One small black spot and faint black pencilings on pleura above base of each leg; much less prominent than in *rubidus*.

Fore femur brownish yellow; tibia yellow, tarsus yellowish white; tip of tibia dusky; median purplish spot on femur, no other markings, other than a very small dark mark on each coxa. Middle and hind legs yellow; tarsi shaded with pale brown, joinings darker. Hind tibia about $\frac{3}{4}$ as long as femur. Wings hyaline; faint milky cloud in stigmatic area. Humeral cross vein deep purplish black in posterior half, white next to costa. Longitudinal veins in anterior and apical portions of fore wing pale yellow, all other veins pale, practically invisible. Abdomen yellowish white. Short dark marks on median line of each tergite, as in *rubidus*; posterior margins of tergites faintly shaded with dark grey, in median area only. No oblique, lateral streaks nor short, dark dashes near spiracles; no darker shading on tergites. Apical segments yellow. Tails smoky at base, pale distally. Genitalia very similar to *rubidus*.

Female imago (alcoholic specimen). Body 12 mm.; wing 14 mm. Similar to male, except as indicated. Small dark dot on median line of pronotum, near to middle of sclerite; lateral margins of pronotum very faintly greyish. All legs similar to middle and hind legs of male; tarsi concolorous with tibiae, joinings slightly darkened. Longitudinal veins of fore wing very pale yellowish brown except in anal region; cross veins in apical portion similar, distinct; elsewhere pale, invisible. About 10 stigmatic cross veins; 3 between bulla and stigma, basal costals very faint. Two dark dots on coxae of middle and hind legs, one only on fore coxa.

Nymph. Described previously, as *Iron* sp. No. 2. A comparison of the postero-lateral spines on segments 6-7 of nymphs of *rubidus* and *subpallidus* is shown in figs. 18 and 19. This nymph is unique among others of the *humeralis* group, because of the relative shortness of these spines, which do not much exceed in length the postero-lateral spines of nymphs of the *longimanus* group.

Holotype—Male imago, reared. Cedar Creek near Glenville, N. C., June 20, 1936. No. 1457.1 in Cornell University Collection.

Allotype—Female imago, reared. Same locality, June 21, 1936. No. 1457.2 in C. U. Collection.

Paratypes—One female imago, reared. Same locality, June 16, 1936. Two female imagos, reared; in same locality, July 10, 1930. No. 1457.3-5 in C. U. Collection.

Iron dispar Trav.

Several nymphs of this species collected at Valle Crucis were reared, and imagos of both sexes obtained. No imagos were taken in flight. Imagos: Valle Crucis, May 25-June 10; and Cherryfield, June 25. Nymphs: Valle Crucis, during same period (J. R. T.), and in autumn of 1934 and 1935 (L. C. Thomsen); Foscue, May 25; Forney's Creek, Aug. 26, 1931 (J. G. Needham); Deep Creek, Aug. 25, 1931 (J. G. N.); Twenty Mile Creek, Apl. 3, 1934 (J. G. N.). The last three localities are in the Great Smoky National Park area. The nymphs from Forney's Creek were previously included in the vial with specimens of *Iron* sp. No. 4.

Iron sp. No. 4

Only two specimens of this species, from Forney's Creek, Aug. 26, 1931 (J. G. Needham). These nymphs are quite similar to *I. dispar*, but distinguished from that species by the dark red-brown abdominal tergites and the distinctive reddish brown shading on the abdominal sternites.

Iron confusus Trav.

This species, a member of the *longimanus* group, has not been previously recorded from North Carolina. Imagos agree well with the type specimens taken in the vicinity of Ithaca, N. Y. Nymphs vary from the New York forms as follows: (1) Ventral markings of last three abdominal segments usually more extensive, consisting of lateral streaks, a pair of submedian spots near anterior margin, and two smaller dots posterior to these; (2) anterior lobes of first pair of gills may be more

prolonged, and so held by the nymph as to be approximated beneath the body; (3) median line of short, fuzzy hairs on abdominal tergites more evident. (Such hairs, usually short and inconspicuous, occur on many Iron nymphs, but are much shorter and weaker than in nymphs of the genus *Ironodes*.)

There is a possibility that the species *I. confusus* Trav. is synonymous with *I. fragilis* Morg.; in that case, however, the size limits of *fragilis* would have to be considerably extended. The only *bona fide* nymphs of *fragilis* that are in the Cornell collection are in such poor condition that it is not possible to be certain whether the gills of the first pair actually meet beneath the body, as indicated in Prof. Needham's figure.* None of the imagos of this species which were reared by Dr. Morgan are now in existence. Nymphs of *confusus* agree well in size with the nymph described by Prof. Needham as *Iron* sp. (8 to 10 mm., depending on the sex of the nymph; he states 9 mm.). However, the imago of *fragilis*, on which the description of the species is based, is stated to have measured but 7 mm., wing 7 mm.; *confusus* has a body length of 9 to 10 mm., wing 10-11 mm. Until this apparent discrepancy in size can be unraveled, we retain the name *confusus* for the larger forms. It should be noted that the nymphs of *confusus* typically hold the gills of the first pair so that the anterior lobes are not directed toward one another beneath the body. However, it is possible to move them, on a dead nymph, so that the tips almost meet. I note, further, that many specimens of typical *confusus* nymphs do have faint dark spots on the abdominal tergites; an apparent statement to the contrary appears in my key to Iron nymphs (Biology of Mayflies).

Several nymphs of this species were reared, and a few imagos taken in flight. The period of flight is in the early evening, just after sunset. Imagos: Valle Crucis, May 27-June 6; and Banners Elk, May 31-June 8. Nymphs: from above localities; also North Toe River at Minneapolis, June 8; and Heaton, June 3. Other specimens were taken by Prof. Needham from Twenty Mile Creek in the Great Smoky Mts., Apl. 3, 1934.

Iron sp. No. 5

Nymphs of this species were collected in Dutch Creek, Valle Crucis, on Feb. 26, 1936 (L. C. Thomsen). None are fully mature; some appear to be only half grown. Male nymphs, body 10 mm.; female

* 1905—Needham, J. G. (Ephemera in) *Mayflies and Midges of New York*, N. Y. State Mus. Bull. 86: 57, Pl. 7, figs. 6-7.

nymphs, 12-13 mm. In alcohol, the general body color is dark red-brown tinged with olive. Dark markings on occiput between eyes and ocelli, and a V-shaped mark from base of antennae to frontal margin. Indistinct dark scroll-like markings on notum. Base of femur, knee, tip of tibia, and entire tarsus, blackish, on each leg. Pale hatchet-shaped mark on basal half of femur; in head of hatchet, the usual purple spot. Anterior and posterior margins of all tergites narrowly black. Dark, paired, submedian spots usually present on tergites 3-9. Gills brown on outer margin; a white strip occupies remainder of outer half; inner half of each, pale purplish. Gills of first pair may meet beneath body of nymph. Tails red-brown, joinings blackish. Venter marked as in *confusus*.

Other nymphs very similar to the above are from Twenty Mile Creek, Great Smoky Nat. Pk., Apl. 3, 1934 (J. G. Needham). In these nymphs, the hairs on the median line of the abdominal tergites are better developed than in any others of the genus *Iron* which I have studied. Otherwise they agree well with the nymphs from Valle Crucis.

Family BAETIDAE

Subfamily Siphonurinae

Genus *Ameletus* Eaton

No imagos of this genus were collected, nor do I have any nymphs among my specimens. However, many nymphs were collected by Prof. Needham in April 1934, in certain areas of the Great Smoky National Park.

Ameletus sp. No 2

1932—Traver, J. R.—J. Elisha Mitchell Sci. Soc. 47: 199.

Mature nymphs which seem to be of this species are from Twenty Mile Creek and Big Creek in Walnut Flats, Apl. 3-7 (J. G. N.). Many of these specimens are more uniformly red-brown dorsally than is indicated in my previous description. Yellow areas present on all tergites: postero-lateral angles; lateral patches; traces of pale median line basally, on apical tergites. A pair of dark, red-brown, submedian streaks in anterior half of each tergite. Inner margin of gill may appear brownish or both margins may be yellow. In some well-marked specimens, tergites 1-2, 7-8, and 10 are distinctly paler.

Ameletus sp. No. 3

A single immature nymph, Cataloochee Creek, Apl. 6, 1934 (J. G. N.). Smaller than the preceding species,—body 7 mm. Frontal portion of head red-brown, occiput yellow. Pronotum yellow; two brown, submedian streaks. Meso- and metanota yellow with many irregular brown markings. Legs yellow; indistinct broad, median, femoral band; tibia and tarsus brown at base and apex; claw brownish. Abdominal tergites 1-2, and 7-8, yellow; posterior margins of 7-8 brown, also faint submedian streaks. Tergites 3-5 largely brown; lateral margins except posteriorly, antero-lateral areas, and large, rounded, submedian spots, yellow. Tergite 6 largely yellow; posterior margin, median triangle based on posterior margin, and smaller submedian triangles based on anterior margin, dark brown. Tergites 9-10 uniformly dark brown. Gills wholly pale. Venter yellowish; sternite 9 largely brown. Usual blackish band across tails.

Genus **Isonychia** Eaton**Isonychia sadleri** Trav.

Another new state record. The large, dark brown nymphs of this species, strikingly marked with a broad, white, dorsal stripe, were very numerous in the streams of the Valle Crucis region. Several were reared. Most of the imagos, however, were taken during their twilight nuptial dance. Great numbers of both sexes congregated over Crabapple Creek at Valle Crucis, and over a stream near Banners Elk. Often they danced high in the air; again, individuals as well as mating couples drifted down almost to stream level. They were strong, tireless dancers, the first to appear and the last to leave the scene of festivity. Imagos: Valle Crucis, May 27-June 6 (J. R. T., L. C. T.); Banners Elk, June 8. Nymphs: Valle Crucis, same period; Cataloochee Creek, Apl. 26, 1934 (J. G. Needham).

Isonychia notata Trav.

Three bronze-brown nymphs taken in Cove Creek north of Vilas are tentatively referred to this species. The tibial spine is shorter than in the type specimens of *notata*. In size and color pattern, however, there are no discrepancies. Two of these nymphs were transported to Penrose, where they lived until June 25. On this date, as I was preparing to move to Alabama, all nymphs in the rearing cages were killed.

Isonychia sp. No. 1

A single female imago, taken in flight near Davidson River, Pisgah National Forest, June 20. Two or three males were seen also, flying very high, headed for the tops of the tallest trees; none could be captured. By the structure of the subanal plate of the female, this species is a member of the *albomanicata* group.

Female imago (dried). Body 13 mm.; wing 13 mm. Frontal portion of head purplish red. Posterior to ocelli, yellow; red shading in antero-lateral angle and on anterior fourth of median line; posterior margin shaded with black, especially next to eye. Antennae dusky. Pronotum shaded with rose in median area, blackish laterally. Remainder of thorax mahogany red-brown; rose-red markings on pleura. Fore femur and tibia deep purplish brown; tarsus purplish grey. Other legs yellow, claws and last tarsal joint dusky. Venation pale brown. Abdomen deep red, brighter than *I. sadleri*. Dorsum and venter very similar; posterior margins narrowly blackish, anterior margins pale. A faint but discernible paler mid-dorsal stripe. Tails white; joinings reddish, in basal portion.

Subfamily Leptophlebiinae

Genus **Paraleptophlebia** Lestage**Paraleptophlebia guttata** McD.

In the early forenoon of June 19, hundreds of males of *P. guttata* were engaged in their nuptial flight, over the Davidson River and above the adjacent willows. The sunlight glinted on their iridescent wings and silvery white bodies as they flew upwards, to glide slowly down with tails and legs outstretched. This was not a continuous performance. All the dancers rested, at intervals of several minutes, on the willow leaves. Now, scarcely one could be seen in flight; another second, and the air was filled with them. Many males and a few females were captured from this dancing throng. Nymphs and imagos were collected from a tributary of the Davidson River on June 20 (J. R. T., L. C. T.); one nymph, Valle Crucis, June 8.

Paraleptophlebia swannanoa Trav.

Immature nymphs were taken at Valle Crucis, May 27; a stream near Blowing Rock, June 7; Cherryfield, June 17; and a tributary of the Davidson River, June 20. No imagos were obtained.

Paraleptophlebia mollis Etn.

This species has not been known before from North Carolina. The mating flight of *P. mollis* takes place just after sunset. Imagos are

represented among material captured in and near Valle Crucis at twilight, May 27–June 2. Other imagos: Banners Elk, May 31–June 8; Cranberry, June 8; mountain road near Penrose, June 12.

***Paraleptophlebia adoptiva* McD.**

Another species of the genus which is recorded for the first time from the state. Imagos: Valle Crucis, early spring, 1936 (L. C. Thomsen), and June 1. Nymphs: Valle Crucis, May 30–June 7; and Foscue, May 25. Other nymphs were taken by Prof. Needham in the Great Smoky National Park, as follows: Moore's Spring on Gregory Bald, Apl. 17, 1929; Twenty Mile Creek, Apl. 3, 1934; Cataloochee Creek, Apl. 6, and a tributary of this stream, Apl. 7, 1934.

Genus ***Habrophlebiodes*** Ulmer

***Habrophlebiodes betteni* Ndm.**

Another species not reported previously from North Carolina. One male imago was taken at Davidson River, June 20; another, a short distance over the state line into Tennessee, near the little settlement of Trade, Tenn., June 9 (L. C. Thomsen).

Genus ***Blasturus*** Eaton

Two undetermined species of this genus are represented: a small species from Hazel Creek, Great Smoky National Park, Apl. 3, 1934 (J. G. Needham); and larger nymphs from Dutch Creek, Valle Crucis, Feb. 26, 1936 (L. C. Thomsen).

Subfamily ***Baetiscinae***

Genus ***Baetisca*** Walsh

***Baetisca thomsenae** sp. nov.**

Closely allied to *B. carolina*. Wings of imago with less intense orange shading; forceps of male relatively longer and more slender. Lateral spine on thoracic shield of nymph somewhat longer, better developed.

Male imago (dried). Body 10–11 mm.; wing 11–12 mm. Eyes red-purplish black. Frontal portion of head red-brown; ocelli with reddish tinge. Base of antenna brownish, filament yellowish. Thorax dark red-brown. Pronotum and posterior portion of head concealed beneath the large eyes. Yellow shading on anterior and antero-lateral margins of mesonotum, and to a lesser degree along the median strip.

* I take pleasure in naming this species for Dr. Lillian Thomsen, who accompanied me on this collecting trip and captured many of the specimens.

Scutellum and adjacent areas purplish red. Yellow shading anterior to base of fore wing, and a streak anterior to middle leg. At base of each wing and above hind leg, a reddish area. Bifurcate process between bases of fore legs quite prominent. Fore legs amber, tinged with reddish brown; all joinings darker. Middle and hind legs very similar in color, slightly paler, joinings less distinctly darkened. Wings hyaline. Entire costal strip of fore wing strongly amber-tinged; a paler tinge of amber extending along membrane for a short distance beyond the base. Extreme bases of both wings deep reddish orange. In hind wing also, an orange flush on membrane for perhaps half the width of the wing. Humeral cross vein red-brown, paler apically; subcosta and radius deep yellow, other longitudinal veins pale purplish brown. Cross veins hyaline, invisible except in stigmatic and anal areas. About 10 stigmatic cross veins; simple, straight.

Dorsum of abdomen rather bright red-brown. Posterior margins, and a small mark at spiracle, black; faint traces of blackish median line; dorsal 'hump' on tergite 6 black at tip. Pleural fold, and adjacent areas of tergites 1-5, deep, dull purplish; faint indications of same, on tergite 6. Pleural fold paler on tergite 7, becoming yellowish on 9. Tergites 9 and 10 lighter red than preceding segments. Venter tan, with faint reddish or purplish tinge. Posterior margins of all sternites black. Purplish red shading laterally and on anterior margins of basal and middle sternites, most evident on basal ones. Genitalia yellowish brown. Tails red-brown, joinings black. See fig. 6 for appearance of genitalia.

Female imago (dried). Body shrunken; wing 13 mm. Similar to male, except as indicated. Entire head rather bright red-brown; narrow dark line at middle of posterior margin; ocelli rose-red. Thorax rather more distinctly reddish than in male. Pronotum extensively shaded with black. Orange tinge on fore wing more restricted, in basal area, than in male. In hind wing, no orange tinge beyond basal third. Abdominal segments 1-6, both dorsum and venter, duller and more distinctly shaded with purplish; apical segments quite similar. Tails somewhat brighter red-brown.

Male imago (alcoholic specimen). Head and thorax olive brown. Purplish shading on pronotum, and pleura of mesothorax. Sternum tinged with red-brown. Scutellum black-margined laterally. Legs yellowish. Longitudinal veins of both wings, intercalaries in cubital region, and cross veins in anal area of fore wing, red-brown; all other cross veins pale, invisible. Fore wing flushed deeply with orange at

base, and with a fainter stain of same color along costal margin to bulla; in some specimens, entire basal half of wing may be faintly orange-tinged. Hind wing deeply orange at base; in some, wholly pale beyond, in others, entire wing faintly orange-flushed, but much less deeply than in typical *carolina*. Abdomen olive brown, 'posterior margins of all segments purplish. Basal and middle segments shaded with purplish red. Irregular darker dots laterally on tergites 1-6, near pleural fold; on each of the five basal sternites, a larger *black* dot next to pleural fold. Tails yellow, slightly darker at joinings.

Female imago (alcoholic specimen). Body 10½-11 mm.; wing 14-14½ mm. Very similar to male. Head and middle area of pronotum with dusky shading. Much purple shading on mesothoracic pleura. Notum more olive green than in male. Purplish markings on thoracic sternum, at posterior margins of pro- and mesosterna. Wings much as in male, but orange tinge on hind wing confined to base, and to a barely visible discoloration of the anal area. Abdomen orange to reddish brown, due to presence of eggs. Strong purplish tinges as in male; lateral purplish black shading on tergites more prominent. Tails deep yellow; joinings narrowly dark brown.

Subimagos. Wings much as in *B. carolina*; in female, rather darker (more blackish). Amount and exact distribution of dark coloring variable in individuals, as is true also of *carolina*. Hind wing of male shows orange flush only at extreme base (in *carolina*, entire basal two-thirds of wing orange-tinged). Entire body duller and darker than in imagos. Black markings at all leg joinings; on tarsi, outer half of each joining a broad black line. Tails olive to reddish brown.

Nymph. Body of male, 8½-9½ mm.; of female, 10-11 mm. Color very variable, ranging from yellow to blackish brown. Many lines and streaks of fine dark dots mark the thoracic shield. In some specimens, the entire background of thorax and abdomen, dorsum and venter, may be obscured by many fine, dark dots close together. Legs banded. In pale specimens, obscure and incomplete dark shading on tibia and tarsus only. In dark forms, femora also are shaded with dark brown. Typically, a distinct black median streak on abdominal tergites; oblique submedian streaks extending *inward* from anterior margin, and other dark marks nearer the pleural fold. Lateral spine of thoracic shield, and forward extension of gena, yellow, the tips piceous. Ventrally, two pale, median spots on mesosternum; a lateral row of pale spots on each side of abdomen, on basal and middle sternites; ganglionic areas of these segments may be yellowish. On pale specimens, these

pale markings are obscure; transverse bands of small, dark dots occupy the middle portion of each sternite. Tails red-brown.

Holotype—Male imago, pinned. Reared from nymph. Valle Crucis, Crabapple Creek, June 5, 1936 (J. R. T.). No. 1459.1 in Cornell University Collection.

Allotype—Female imago, pinned; reared from nymph. Same data. No. 1459.2 in C. U. Collection.

Paratypes—1 male and 1 female imago, pinned; Valle Crucis, N. C., May 26, June 2, 1936 (J. R. T.): 5 male, 2 female imagos, in alcohol; June 2-14, 1936; Valle Crucis, N. C. Same collector. No. 1459.3-11 in Cornell University Collection.

Differences between this species and *B. carolina* may be summarized as follows. Nymph: (1) anterior extension of gena slightly more upturned; (2) lateral spine of thoracic shield better developed, especially in male, so that the entire shield is relatively wider; (3) claws slightly shorter and more slender; (4) spines on 8th abdominal segment tending to turn downward and inward, rather than upward and outward, as in *carolina*. Imagos: (1) forceps of male longer, and more slender in basal portion; (2) median projections on subanal plate of female shorter and stouter; (3) orange tinge in both wings less intense, may be reduced in extent as well as intensity. In both stages, the size is consistently larger than in specimens of *carolina*. Although all of the differences listed are relative, I consider this to be a good species, and one distinct from the closely-allied *carolina*. See plate for figures showing relative differences between the two species: figs. 9, 10, 11, 12, 14, and 15.

On the morning of May 26, a fine male imago of *Baetisca thomsenae* was resting on the window screen of my room at the Valle Crucis School. It was quickly captured, and became my first specimen of this species. I located a few *Baetisca* nymphs that day in Dutch Creek; some were in the riffles, others among sand and gravel in water over a foot in depth. In the forenoon of June 2, many *Baetisca* nymph sloughs were found clinging to rocks near a ford in Crabapple Creek. So I started to search for the nymphs. Remembering my previous experience in collecting *Baetisca* in the piedmont, I held a hand screen in the current and stirred about among the rocks and gravel upstream. Each time the screen was lifted from the water, it held several *Baetisca* nymphs. I was most curious to discover the exact habitat of these nymphs, in relation to the stones, gravel and sand in the stream bed. Presently, as I rested from collecting, I noticed a slight stir among the fine gravel in the lee of a

rock. It was a *Baetisca* nymph, shifting its location. Looking more closely, I was able to make out the dorsal shields of several other nymphs, each partly buried in the fine gravel or sand, resting with head upstream. If disturbed, a nymph would be carried a short distance downstream by the current, but usually swam slowly upstream again, short tails vibrating. Coming to rest on a stretch of sand, a few rapid kicks served to conceal it again, and anchor it against the push of the current. Whether yellowish or blackish, the nymphs so resembled tiny pebbles in the stream bed that I frequently picked up a pebble, thinking it to be a nymph. A few days later, in a series of riffles below the ford, I watched a *Baetisca* nymph for some time, as it alternately rested and shifted from place to place. *Baetiscas* were found in the same location also, in one of the streams near Banners Elk. They could be picked up by hand readily, once they were located.

Although I watched forenoon, afternoon, and evening, I was unable to see a single imago in flight. None were represented in my collections of specimens taken at sunset each day. Nor were any found resting on vegetation near the stream. Do they emerge very early in the morning, or late at night? And at what hour does the mating flight occur? I do not know. Nymphs in rearing cages usually transform to the subimago stage in the forenoon, and the imago stage is likewise attained between 7 and 11 a.m., in cases observed. Thus it is possible that the nuptial flight takes place between 11 a.m. and 2 p.m.

Subfamily Ephemerellinae

Genus *Ephemerella* Walsh

I. *Bicolor* group

Ephemerella doris Trav.

A single male imago, taken at Valle Crucis on May 30, is placed tentatively in this species. Although it does not conform to the type material in all respects, it is certainly very closely allied to *doris*, if not an actual variant of that species.

Ephemerella funeralis McD.

This species has not been reported previously from North Carolina. Several nymphs are present in material collected by Prof. Needham in the Great Smoky National Park, Apl. 3-7, 1934. Specimens are from Cataloochee Creek and Big Creek at Walnut Flats.

***Ephemerella coxalis* (?) McD.**

Another new state record. Three nymphs, taken at Valle Crucis in late autumn, 1934 (L. C. Thomsen), are so closely allied to *coxalis* that I hold them tentatively under that species.

***Ephemerella temporalis* McD.**

Several nymphs were collected by Prof. Needham from Hazel Creek, Great Smoky National Park, Apl. 4, 1934.

II. *Invaria* group.***Ephemerella dorothea* Ndm.**

This species is of common occurrence throughout the piedmont and Appalachian regions of the state. Several imagos were reared from nymphs taken at Valle Crucis and Heaton, while many others were captured during their evening nuptial flight. There is a considerable size range among individuals of this species, both in nymphs and imagos. Thus, male imagos collected during 1936 vary in length of body from 6 to 8 mm. Females vary also as regards the amount of ruddy coloration on head, thorax, and abdomen. Some are wholly pale except for a black line on anterior margin of pronotum, and dark posterior margins on the basal and middle abdominal segments. In small females from the Davidson River, the head is quite bright red; entire dorsum of thorax and abdomen ruddy brown, only the lateral margins of the apical tergites creamy; ruddy shading on posterior portion of prosternum; fore femur distinctly reddish. Males are more uniformly creamy white; eyes ruby-red; posterior margins of basal and middle abdominal tergites may be faintly smoky. In a small male from Davidson River, however, the dusky bands on the tergites are more prominent than usual; a longitudinal red streak is present on each side of tergites 5 and 6, traces of same on 4 and 7; middle area of tergites 8-9 rather dark red-brown. In both sexes, the tails of the imagos may exhibit faint reddish rings at the basal joinings. Nymphs from the vicinity of Valle Crucis and Heaton have more dark markings dorsally than the more uniformly colored specimens from the piedmont. These dark specimens have a pale band on the head between the antennae; pale lateral angles on pronotum, pale paired spots on posterior margins of tergites.

Imagos: Valle Crucis, May 27-June 7, Heaton, June 3, Davidson river, June 11-20 (L. C. T., J. R. T.). Nymphs: Valle Crucis, same period; North Toe River, June 8; and Heaton, June 3.

***Ephemerella dorothea*—Variety A.**

Several imago captured near Banners Elk on May 31 are so similar in general appearance to typical specimens of *dorothea* from Valle Crucis and Heaton, that I am unable to distinguish them except on genitalic structure. The penes of the Banners Elk males bear an unusual number of spines, 12 or 13 on each side (see fig. 3); the usual number of spines for this species is 6 to 9 on each side, typically 7 or 8. Unfortunately, all of these specimens were taken in flight, none were reared. All reared males from Valle Crucis and Heaton are normal as to number of spines on the penes. Nymphs collected at Banners Elk are similar to many of those from Valle Crucis. Therefore I am holding the Banners Elk specimens as a variety, perhaps an aberrant form, of *dorothea*.

***Ephemerella rotunda* Morg.**

Many nymphs of this species were collected by Prof. Needham, Apl. 3-6, 1934, in the Great Smoky National Park, from Cataloochee Creek and Twenty Mile Creek.

***Ephemerella fratercula* (?) McD.**

This species has not been reported hitherto from the state. A single male imago was taken in flight at Valle Crucis, on June 5. As this may prove to be a new species, closely allied to *fratercula*, I present a description of the single specimen, which is preserved in alcohol.

Male imago. Wing 8 mm. Eyes bright orange. Frontal portion of head yellow, vertex olive brown. Pronotum yellow, median area shaded with brown. Mesonotum light reddish brown, rather sharply contrasted with the yellowish pleura and sternum. Faint brownish markings along lateral margins of mesonotum; tip of scutellum creamy white; median anterior area with brownish shading. Metanotum yellow, posterior margin dark brown. Fore legs missing. Middle and hind legs whitish, tarsi very faintly smoky; tarsal joinings narrowly pale brown. Wings hyaline. Longitudinal veins of both wings very pale brown, cross veins invisible; costal margin silvery white except at apex. Dorsum of abdomen smoky to olive brown; posterior margins of basal tergites darker. Basal and middle segments semihyaline, apical segments opaque. Venter much paler than dorsum, but with faint smoky tinge; posterior margins of basal sternites deeper smoky. Tails pale smoky at base, becoming whitish distally; not darkened at joinings. Genitalia shown in fig. 2.

The spining of the genitalia, as well as the general coloring, resembles

fratercula so closely that I am holding the single specimen under that name. I would call attention to the following differences, however. Tails *not* dark-ringed, as in typical *fratercula*; venation pale brown instead of hyaline; no mid-ventral ganglionic markings, as in that species.

III. *Needhami* group.

Ephemerella septentrionalis McD.

Another species not reported hitherto from North Carolina. One male and three female imagoes were taken in flight at Valle Crucis, May 25-27. No nymphs were collected. The extraordinarily long legs of this species, both in the nymph and the imago stage, are distinctive.

Ephemerella catawba Trav.

Many nymphs were collected at Valle Crucis, and in the Watauga River between Valle Crucis and Vilas. A female imago and a subimago were reared; two other imagoes were taken in flight. No males were obtained. The female has not been described heretofore.

Female imago (alcoholic specimen). Body $8\frac{1}{2}$ mm.; wing $9\frac{1}{2}$ mm. Head and thorax greenish yellow; remainder of thorax deeper yellow. A low carina on pronotum, its anterior projection faintly dusky. Margins of mesonotum, and submedian triangular areas anterior to scutellum, dark brown. Posterior portion of mesosternum brownish. Legs yellow; tarsal joinings faintly darkened; claws dusky. Wings hyaline; stigmatic area faintly milky. Costa, subcosta, and radius of fore wing yellowish, all other veins paler, whitish; 8 to 9 stigmatic cross veins, wholly pale, anastomosed near costal margin, somewhat aslant. Abdomen discolored, due to presence of eggs. Pleural fold pale; sternites appear to be paler than tergites. Posterior margins of tergites narrowly darkened; those of sternites subhyaline, pale. Tails yellowish white, pale brown at extreme base; all joinings distinctly black-ringed.

Among my pinned specimens (Valle Crucis, May 25) are two females which I take to be *E. catawba*. Note differences between alcoholic and dried specimens, as to color of head and thorax.

Female imago (dried). Frontal portion of head yellow, antennae dusky, vertex and occiput pale red. Pronotum red-brown, with blackish shading on the low median carina. Mesonotum light red-brown; submedian triangles anterior to scutellum darker. Pleura and sternum flesh-colored, with many creamy markings. Legs yellow. Fore femur faintly flushed with pink, somewhat dusky at apex. Apical margins

of tarsal joinings on all legs faintly dusky. Dorsum of abdomen reddish brown with olive tinge; posterior margins blackish. Tergite 10 rather brighter red-brown. Sternites 1-6, and 9, yellow; 7 and 8 shaded with reddish. Tails as in alcoholic specimens.

Allotype—Female imago, reared. Watauga River near Valle Crucis, May 29, 1936 (J. R. T.). No. 1094.2 in Cornell University Collection.

Other specimens (nymphs): Great Smoky National Park, June 11, 1931 (J. G. Needham); Conestee Creek, June 20, (L. C. Thomsen, J. R. T.); North Toe River at Minneapolis, June 8 (L. C. T.); Linville River at Linville Falls, June 2 (L. C. T., J. R. T.); twenty Mile Creek, Great Smokies, Apl. 3, 1934 (J. G. N.); Banners Elk, June 3.

Ephemerella sp. No. 3

A single nymph, taken in Twenty Mile Creek, Great Smoky National Park, Apl. 3, 1934 (J. G. Needham), is intermediate in its characters between *E. catawba* and *E. rotunda*. General color, dark red-brown. Paler spots on head, anterior to each eye; occiput indistinctly mottled. Entire lateral margin of pronotum yellow. Meso- and metanota unmarked. Tibiae and tarsi dark-banded. Spines on fore femur mainly along posterior margin; do not form a continuous band across dorsal surface. A pair of short, submedian spines on posterior margins of tergites 3-8. No pale markings on abdomen. Whorls of spines at tail joinings prominent in basal half. Body (male nymph) $8\frac{1}{2}$ mm. A quite similar nymph, from Cataloochee Creek, Apl. 6, 1934 (J. G. N.), has a broad, pale, dorsal band the entire length of the body, as in one color phase of *E. needhami*.

Ephemerella sp. No. 4

Allied to *E. aurivillii* but larger; body (female nymph) 10-12 mm. Dorsal abdominal spines much better developed. General color red-brown. Two submedian, small, white spots on frontal margin of head; a large, rectangular, white spot anterior to middle ocellus; a smaller pale spot laterad of lateral ocellus. Occiput and entire dorsum of thorax mottled with darker blotches. Legs pale red-brown. Femora mottled with yellowish blotches; on fore femur, a small cluster of spines near posterior margin, but not forming a band across dorsal surface. Inner apical angle (tibial spine) of fore tibia slightly produced. Neither tibiae nor tarsi dark-banded. Claws with many pectinations, as in *E. catawba*. Pale submedian blotches may be present on tergites 5 and 6, next to inner margin of gills; on outer edge of this pale blotch, a deeper red streak. Postero-lateral extensions and spines yellowish;

no dark transverse band. Dorsal abdominal spines well developed; present on tergites 1-9, longest on 4-7. A series of short spines or tubercles extends from base of each spine forward almost to anterior margin of tergite. Tails yellowish red, darker apically; whorls of very short spines at joinings in basal half. Paler ventrally, posterior margins of sternites narrowly darkened; indistinct lateral line of marks near pleural fold. No mature male nymphs.

Ephemerella sp. No. 5

A small species of the *needhami* group, allied to *E. catawba*. Body of mature female nymph, 6-6½ mm., of male, 5-5½ mm. General color light red-brown; dorsum of abdomen with some purplish shading. Pale marks on head: anterior to median ocellus; anterior to eyes; submedian spots on posterior margin; epicranial suture. Lateral margins of pronotum pale; also irregular lateral markings. Mesonotum with irregular dark brown markings between and anterior to wing roots. Legs pale red-brown; no dark bands. Spines on fore femur numerous before apex, but not forming a continuous band. Postero-lateral extensions of abdomen well developed. Paired, submedian, dorsal spines on tergites 4-8; anterior to each spine, a series of small tubercles. Posterior margins of tergites purplish black; lateral areas shaded with purplish; middle segments may be white-blotched next to gills. Tails wholly reddish brown; whorls of spines at joinings in basal half. Ventrally, traces of a lateral line of dark dashes, one pair to a sternite, nearer to pleural fold.

IV. *Fuscata* group.

***Ephemerella cornutella* McD.**

Nymphs of this species in my collection are from Valle Crucis, May 27-June 8; Heaton, June 3; stream near Blowing Rock, June 7; Cove Creek near Vilas, June 9; Davidson River, June 20; Lecky Gap, June 25; and Little River, Great Smoky National Park, June 26.

***Ephemerella cornuta* Morg.**

Imagos: Lecky Gap, June 16; Valle Crucis, June 10. Nymphs: Heaton, June 3; North Toe River, June 8; Cove Creek, June 21; Valle Crucis, May 27-June 10.

***Ephemerella longicornis* Trav.**

A few immature nymphs from the type locality, Cedar Creek near Glenville, June 15, and one nymph from a stream near Banners Elk, June 3.

***Ephemerella lata* Morg.**

One nymph, Valle Crucis, May 28.

***Ephemerella cherokee* sp. nov.**

E. tuberculata Morg., as treated in Traver, J. Elisha Mitchell Sci. Soc. 47: 175. 1932.

Nymphs and imagoes from North Carolina, which I had previously considered to be *E. tuberculata* Morg., do not agree in many respects with Canadian specimens which Dr. McDunnough has reared. The type material, a single nymph taken near Ithaca, is no longer in existence. Dr. Morgan's figures indicate a tibial spine even longer than in the Canadian forms; occipital tubercles large, slightly convergent apically. The description calls for a "double row of distinct brown spots" ventrally. As the Canadian specimens seem nearer to the original description than do those from North Carolina, I am considering the latter as representing a new species.

Imago (alcoholic specimens). Body of male, 10 mm.; female, 9-10 mm.; wing of male, 10 mm.; female, 10-10½ mm. Distinguished from the Canadian specimens (on which the description in *Biology of Mayflies* is largely based) by the much darker venation, and the entire lack of dark ventral markings on the abdomen of the female. All longitudinal veins yellowish at extreme base, distinctly red-brown beyond. Cross veins in stigmatic area and in adjacent subcostal space pale red-brown; all others very pale, invisible except in apical area of radius of fore wing, and subcostal space of hind wing. Those of stigmatic area strongly anastomosed near costal margin. Eyes of male smaller in the North Carolina form; notum and sternum of thorax darker, blackish brown. No dark lateral dash on each sternite, next to pleural fold; other ventral markings in male diffuse, most evident on apical segments, obsolescent on basal ones. In female, no dark markings ventrally.

Nymph. Size very slightly larger than Canadian specimens. General color of body light red-brown; in life, greenish, often with a prominent whitish mid-dorsal stripe. Somewhat less hairy than Canadian form. Rather fewer tubercles on dorsal surfaces of femora, especially noticeable on the second leg. Tibia of second leg fully as long or slightly longer than femur; in Canadian forms, slightly shorter. Tibial spine of fore leg extends forward only about ½ the length of the tarsus, while in Canadian specimens it extends almost half the length of the tarsus. Tubercle on lateral margin of pronotum much smaller than in Canadian

form; submedian tubercles on anterior margin of mesonotum, and median tubercle between wing cases, lower and more rounded. Paired submedian spines on abdominal tergites 3-7 relatively shorter. Lateral margin of 8th segment practically straight, the spine slightly incurved. In Canadian specimens, this margin is convex in basal portion, slightly flaring beyond, somewhat as in *E. allegheniensis*; spine barely or not at all incurved. Ventral abdominal markings much less distinct; when evident, reddish brown. Canadian specimens have a row of prominent blackish lateral streaks on each side.

Holotype—Male imago, reared. Cedar Creek near Glenville, N. C., July 25, 1930 (J. R. T.). No. 1460.1 in Cornell University Collection.

Allotype—Female imago, reared. Ocona Lufty River near Cherokee, N. C., June 28, 1930 (J. R. T.). No. 1460.2 in C. U. Collection.

Paratypes—Two female imagos, reared. Conestee Creek near Cedar Mt., N. C., July 19, 1930 (J. R. T.). No. 1460.3-4 in C. U. Collection.

Ephemerella sp.

Many female imagos were ovipositing in the Davidson River in the early forenoon of June 5, and again on June 16. They came flying steadily upstream toward the riffles where we were collecting, two or three to a dozen at a time. Occasionally one would dip down toward the surface of the water. They were unaccompanied by males.

The sprinkling of black dots on the femora indicates a relationship to the *fuscata-tuberculata* division of this group. There are no traces of black ventral markings on the abdomen, as in *tuberculata*; size smaller and venation paler than in *cherokee*; body color and venation darker than in *fuscata*, the red vertex of that species replaced by brownish black. Specimens seem too large to be *wayah*, imagos of which are not known.

Female imago (alcoholic specimen). Body 6-7 mm.; wing 8-9 mm. Frontal portion of head dark red-brown; vertex and occiput blackish, as is the pronotum. Thorax deep blackish brown, pleura somewhat paler. Venation light red-brown, longitudinal veins distinct. Fore legs blackish brown; femora of middle and hind legs olive brown, tibiae and tarsi yellowish; all femora with sprinkling of small black dots. Brownish shading at extreme base of both wings. Abdomen dark olive brown with reddish tinge; slightly paler ventrally. No apparent darker markings. Tails deep smoky brown, narrowly pale at joinings. Pinned specimens very similar. Specimens taken on June 16 are slightly smaller than those of June 5, otherwise similar.

V. *Serrata* group.***Ephemerella deficiens* Morg.**

Imagos: Banners Elk, June 8; near Penrose, June 12; Davidson River, June 19-20. Nymphs: Valle Crucis, May 27-June 8; Heaton, June 3; Davidson River, June 17; Lecky Gap, June 16.

VI. *Simplex* group***Ephemerella* sp. No. 1**

A single specimen of this species, a mature female nymph, was taken in a stream near Blowing Rock on June 7 (L. C. Thomsen). Body $6\frac{1}{2}$ mm. in length. Femora definitely wider than in *E. simplex*, to which it is allied; lateral margins of abdomen likewise more expanded, so that body appears wider. Tails distinctively marked: except at base, a series of three or four brownish joints alternate with one pale joint; joinings darker on brown areas. This appears to be a distinct species.

Undetermined female imagos

Among the specimens taken in flight in and near Valle Crucis are a dozen or more female imagos, representatives of four or five species. Some are evidently of the *invaria* or the *needhami* group, others seem to be of *serrata* or *simplex* group. I am unable to determine these as to species at the present time.

Subfamily Baetinae

Genus *Callibaetis* Eaton***Callibaetis* sp.**

Two nymphs of an undetermined species of this genus were taken at Valle Crucis in the autumn of 1934 (L. C. Thomsen).

Genus *Baetis* Leach***Baetis incertans* McD.**

A single male imago was reared from nymph; Valle Crucis, May 30. Nymphs: Valle Crucis, June 8; Cove Creek north of Vilas, June 9.

Nymph. Body of male, $4\frac{1}{2}$ mm. Head and thorax red-brown. Legs dark-banded. Abdominal tergites 1-3 and 6-8 largely brown; lateral margins and middle line at anterior margin pale, on 1-3. Median and lateral areas of tergite 4 pale, remainder brown. Tergite 5 largely pale, except for two dark submedian dots and narrow dark antero-lateral streak. Tergites 9-10 pale, unmarked. A pair of obscure, dark, submedian dots on tergites 6-8. Sternites 6-8 faintly dark-shaded;

posterior margins narrowly dark. Gills obovate; white, margins narrowly brown, main trachea visible in basal half only. Tails pale, tips darker.

Baetis cingulatus (?) McD.

A single male imago taken in a spider's web near Trade, Tenn., June 9 (L. C. Thomsen). This species has not been reported previously, either from North Carolina or Tennessee.

Baetis sp.

Several female imagos, caught in a spider's web at Conestee Creek, above the town of Cedar Mountain, June 28.

Two species of nymphs from Valle Crucis, May 26 and June 8. One species from Cove Creek at Sugar Grove, north of Vilas, June 9. One species, Twenty Mile Creek, Apl. 3, 1934 (J. G. Needham).

Genus Acentrella Bengtsson

Further study of the specialized Baetine group, Neotropical and Palaearctic as well as Nearctic, has convinced me that many genera are involved, and that it may not be possible in all cases to be certain of the genus unless both the nymphs and imagos are known. The nymph of Bengtsson's genus *Acentrella* is distinct from all species of *Baetis* except those allied to *bicaudatus* Dodds, by reason of its two-tailed condition. In the imago, the costal angulation of the hind wing is wholly wanting; a rounded or somewhat truncate 'penis-cover' is present on the male genitalia, between the bases of the forceps. Given both nymph and imago, it is possible to separate a certain group of *Baetis*-like species from all others in the subfamily; it is to members of this group that Bengtsson's generic name applies. I am convinced now that this group is worthy of generic rank, as I have indicated in *Mayflies of North Carolina*, although the species are treated under the genus *Baetis*, in the *Biology of Mayflies*.

Acentrella ampla Trav.

A small, rounded penis-cover is present between the forceps cases, in this species. The original figure merely suggests the presence of this structure; it was, in fact, not clearly indicated until the genitalia had been treated in caustic potash and re-mounted.

Genus Centroptilum Eaton

Centroptilum sp.

Females of two undetermined species of *Centroptilum* were taken in flight. One species, in which the body is rather uniformly reddish

brown, venter of abdomen paler, is represented by specimens taken at Banners Elk, May 31, and Cranberry, June 8. A single female of the second species, smaller and yellow-brown, was taken in the Pisgah National Forest on June 20.

Genus *Pseudocloeon* Klapalek

Pseudocloeon carolina Bks.

A single nymph, Valle Crucis, June 6.

Pseudocloeon dubium Walsh

This species is represented by nymphs from Cove Creek, June 9; stream near Blowing Rock, June 7 (L. C. T.); and the Great Smoky National Park, June 11, 1931 (J. G. Needham).

Pseudocloeon sp.

Undetermined female imagos of this genus were taken in flight at Davidson River, June 20; Penrose, June 12; near Blowing Rock, June 7; Conestee Creek, June 22.

Several nymphs of the *carolina-cingulatum* group are from Valle Crucis, May 25; Banners Elk, May 31; and Heaton, June 3. These resemble *carolina* in general coloration, but are more slender.

Genus *Cloeon* Leach

Cloeon sp.

A single undetermined female imago, Valle Crucis, June 7.

ALABAMA*

In Alabama, Tuscaloosa was selected as headquarters for collecting trips and for the rearing of nymphs. Tanks of running water, in the Zoology Department of the University of Alabama, were made available to us for rearing purposes, thanks to the kindness of Dr. Septima Smith of the Zoology Department of that institution.

* Small streams in northern and central Alabama, in which we made collections, are relatively poor in mayfly, stonefly, and caddisfly faunas. This paucity of the mayfly nymphs in the small streams was very marked, in contrast with the rich collecting we had just left, in the mountains of North Carolina. It was also in striking contrast to the wealth of material to be obtained in the Odonate group, both dragonflies and damselflies. In only one stream of a dozen or more in which we collected assiduously, did we find more than six or eight mayfly nymphs, and that after an hour or more of endeavor. The larger rivers yield representatives of the Ephemeridae, as well as a few species of the other two families. No attempt was made to collect in the Black Warrior River in or near Tuscaloosa; it appears to be much polluted as it passes through that city.

All species listed here, with the exception of *Hexagenia bilineata*, have not been reported previously from the state of Alabama.

Family EPHEMERIDAE

Subfamily Ephemerinae

Genus *Hexagenia* Walsh

***Hexagenia bilineata* Say**

Imagos of both sexes were numerous on the store windows in Tuscaloosa on the evening of July 2. On July 4, many imagos and subimagos were seen on a bridge over a river between Birmingham and Decatur. Many were tangled in spiders' webs. Great numbers of nymphal skins were floating on the surface of the water, but we were unable to obtain any of them. The following day, many imagos were taken from low vegetation along the shore of Wilson Lake, just above the dam at Muscle Shoals.

***Hexagenia orlando* Trav.**

Three male imagos taken in Tuscaloosa, June 29–July 2, correspond quite closely to specimens of *orlando* taken at Spring Creek, Ga. The type specimen of this species is paler, due to immersion in alcohol for a longer period of time, but is similar in essential features. In the Alabama specimens, the middle and hind legs are wholly yellow except for claws and distal tarsal joints, which are black; in typical *orlando*, all joinings of these legs are brown. Four females taken along with the males are evidently of the same species. Another female accompanying them has the dark markings on dorsum of abdomen much reduced; it may be only a very pale form of the same species. Nymphs of this species are not known.

***Hexagenia* sp.**

Nymph (described from specimens taken at Cooley Creek). Body (including tusks): female, 30 mm.; male, 22 mm. Mandibular tusks as in fig. 17. Frontal process of head rounded laterally, anterior margin flattened. Head and thorax reddish brown. Legs yellow, with usual fringes of dark orange hairs. Abdomen yellowish white. A wide, grey, mid-dorsal band encloses pale, broken, submedian lines; from each end of this dark band, on the posterior margin, an oblique grey streak extends antero-laterally. Gills and gill fringes deep purplish grey. Tails white to yellowish, fringed with yellow hairs. Ventrally, traces of purplish transverse dashes at postero-lateral margins, and in

median area of sternites 7 and 8. In male, black, submedian, transverse dashes on anterior margins of basal and middle sternites.

Specimens were taken at Cooley Creek, between Tuscaloosa and Birmingham, July 4. An immature nymph was taken from Big Sandy Creek near Coaley, on July 2 (R. E. Hodges); nymph skins of the same species floated near the shore of Thompson's Lake, near Tuscaloosa, on June 29. Unfortunately, no adults were reared, nor were any found in the same vicinity as the nymphs. Can this be the nymph of *H. orlando*? It is evidently a species whose habitat is small lakes and streams, rather than the large rivers usually inhabited by *H. bilineata*.

Genus *Pentagenia* Walsh

Pentagenia vittigera Walsh

Two females were captured from store windows in Sheffield, on the evening of July 4.

Family HEPTAGENIIDAE

Genus *Stenonema* Trav.

Stenonema interpunctatum Say

Imagos of this or a closely allied species were taken from vegetation on the shore of Wilson Lake, on July 5. A single female from the North River near Tuscaloosa may be of this same species.

**Stenonema smithae* sp. nov.

A species of the *pulchellum* group; superficially very similar to *S. integrum* McD., but differing in details of genitalic structure.

Male imago (dried). Body $7\frac{1}{2}$ mm.; wing $7\frac{1}{2}$ –8 mm. Head pale. A faint ruddy transverse band across median carina. Tip of basal segment of antenna, and filament, dusky. Eyes large; piceous. Thorax pale yellowish white; mesonotum, also areas on pleura, flesh-colored. Scutellum white. Fore femur and tibia yellow; ruddy median and apical bands on femur; tip of tibia black. Tarsus paler, yellowish white; claws and tip of distal joint, and all joinings, dusky. Middle and hind legs yellowish white; ruddy bands on femora, as in fore leg; tips of tibiae, claws and distal tarsal joints, dusky. Basal joint of fore tarsus less than half the length of the second. Wings hyaline. Venation of fore wing brown, rather darker than in *integrum*; subcosta and radius yellowish.

* I take pleasure in naming this species for Dr. Septima Smith, who extended to us many courtesies during our stay in Alabama.

low; all longitudinal veins finer than cross veins. Humeral cross vein infuscated, also base of subcosta. Cross veins more regularly distributed than in *integrum*,—less tendency toward serial arrangement. Usually three costal veins before the bulla. Stigmatic cross veins 7 or 8 in number; simple, straight. Very faint reddish tinge in stigma. Venation of hind wing wholly pale; outer margin very narrowly dusky in apical portion only.

Abdomen pale yellowish. Stigmatic dots well developed. Posterior margins of tergites narrowly dark, most evident in median area. Apical tergites faintly flesh-colored. Pleural fold yellow. Sternites unmarked. Tails whitish, alternate joinings narrowly purplish black. Genitalia more distinctly L-shaped than is the case in *integrum*.

Male imago (alcoholic specimen). Dusky markings at middle of anterior margin of mesonotum, and along its antero-lateral angle. A faint dusky streak on prothorax, above leg base; on mesothoracic pleura, very faint dusky pencilings above leg bases. Fore femur with faint ruddy tinge; apical ruddy bands of all femora more distinct and more reddish than median ones. Abdominal segments 1-7 whitish, not yellowish as in dried specimen.

Female imago (alcoholic specimen). Body 7½-8 mm.; wing 8-9 mm. Similar to male except as indicated. Reddish stigmatic tinge may be more evident than in male, or it may be wholly obsolescent. At bulla, cross veins in first three spaces are: 1, 2, 2. . Cross veins somewhat more numerous than in male; tend to be arranged irregularly in series. A blackish longitudinal penciling extends the length of each femur, near middle of outer surface. A faint reddish tinge may be present on basal third of each tibia. Dusky pencilings on thoracic pleura, spiracular dots, and dark posterior margins of abdominal tergites, more evident than in male. Thorax and apical abdominal segments more yellowish. Lateral tracheae dusky, on apical segments. Dusky margin of hind wing more extensive.

Nymph. Head dark brown; frontal portion thickly freckled with small pale dots. Pale area along outer margin of eye; one dark spot in this pale border. A fleur-de-lis shaped pale mark anterior to median ocellus. Pale areas next to inner margin of eye, and a diamond-shaped pale median area between eyes. Bases of antennae dark brown, filament paler. Thorax dark red-brown; pronotum darker and less red than mesonotum. Pale markings on pronotum: narrow median line; a pair of submedian dots; lateral triangular marks; middle portion of lateral margin, and an inwardly-directed anterior arm from same.

Median line of mesonotum pale; a few small and indistinct pale dots anterior to wing roots. Femur largely dark except at base, apex, a pre-apical band, and small pale median areas enclosed by dark color. Near apex, a reddish band as in imago. Tibia yellow; a narrow basal and a wider median dark brown band. Apical half of tarsus pale.

Abdominal tergites 1-5 pale laterally, and irregularly in median area, elsewhere dark brown. Tergites 6-10 largely dark brown; a pale lateral triangle on 6 and 7; anterior margins of 9 and 10, and submedian dashes from anterior margin of 9, likewise pale. Venter yellowish. Brown streaks along lateral margins of sternite 9. Very faint indications of dark submedian dashes from anterior margin of each sternite, and a dark dot near end of each. On sternite 9, a faint dark median spot at anterior margin; a transverse dark line extends laterad from it. Tails red-brown at base, followed by a yellow area, in which joinings are brown; in apical two-thirds, three dark joints alternate with one pale joint. Gills purplish grey; tracheae black.

Holotype—Male imago, pinned. Reared from nymph. Spencers Mill, Tuscaloosa, Ala., July 1, 1936 (J. R. T.) No. 1461.1 in Cornell University Collection.

Allotype—Female imago, in alcohol; reared. Same locality, July 3, 1936 (J. R. T., S. C. Smith). No. 1461.2 in C. U. Collection.

Paratypes—Three male imagos, two female imagos; same locality, June 30—July 8, 1936 (J. R. T., S. C. Smith). No. 1461.3-7 in C. U. Collection.

Nymphs of this species were collected at Spencers Mill near Tuscaloosa, Ala., and reared in tanks in the Zoology Laboratory at the Univ. of Alabama. The imagos differ from the allied species *S. integrum* McD. as follows: (1) Larger size; (2) genitalia of male distinctly L-shaped; (3) cross veins in wings less regularly arranged in series; (4) spiracles marked by a distinct dark dot rather than a dash; (5) no median dorsal markings on tergites.

***Stenonema alabamae* sp. nov.**

Another species of the *pulchellum* group, allied to *S. integrum* McD. Basal joint of fore tarsus longer than in any known species of this genus.

Male imago (dried). Body 6 mm.; wing 7 mm. Frontal portion of head pale; a wide, dark, transverse band across median carina. Much ruddy shading on vertex and occiput. Pronotum clay-colored, median areas dusky. Pleura and sternum of mesothorax flesh-or-clay-colored; alabaster white areas on pleura, and on sternum between middle legs.

Anterior to wing roots, and on pleura, distinct ruddy shading. Blackish pencilings above leg bases. Mesonotum blackish brown; tip of scutellum, and small area anterior to it, whitish. Fore legs yellowish white; femora faintly smoky. Median and apical ruddy bands on femora; darker shading at base. Tips of tibiae blackish. Claws, most of distal joints of tarsi, and all joinings, dusky. Basal joint of fore tarsus *fully three-fourths as long as the second*. This is true of both legs. Is it an abnormality? On this character, the species would fall into *Cinygmula*; but genitalia, venation, and all other characters are that of the genus *Stenonema*.

Middle and hind legs whitish; femora twice-banded with reddish; tibial and tarsal joinings, claws and tips of tarsi, dusky. Wings hyaline. No crowding of cross veins at bulla; in first three spaces, these are: 1, 2, 2. Base of costa and subcosta, and humeral cross vein, infuscated. Three main veins of costal margin yellow, well developed; other longitudinals much finer than cross veins, brown. Cross veins blackish brown; some tendency toward serial arrangement as in *S. integrum*. Five costals before bulla; seven or eight in stigmatic space; simple, may be slightly aslant. Distinct reddish stigmatic stain; seems wholly confined to subcostal space. A wide smoky border on hind wing occupies fully one-fourth of the width of the wing. Abdominal segments 1-7 semihyaline. Wholly white ventrally; tergites with distinct smoky tinge. A rather wide dark brown posterior border on all tergites; spiracular dots present, but more or less obscured by the dark margins. Segments 8-10 opaque. Tergites 8-10 rather bright red-brown; a pale median spot on tergite 10. Sternites whitish. Tails whitish; alternate joinings purplish brown. Genitalia much as in *S. rubrum* McD.

Female imago (dried). Body 6-7 mm.; wing 8 mm.

Head whitish; band on carina usually present, as in male. Thorax flesh-colored; sternum whiter than pleura. Fore leg pale yellowish, other legs whitish; marked as in male, the femoral bands purplish red. All longitudinal veins yellowish, fine; cross veins very heavy, dark brown; as in male, a tendency for these to be arranged in a series, but the wide interspaces thus formed appear *before* the bulla (in *integrum*, these are *beyond* the bulla). At bulla, cross veins may be: 1, 2, 2; or 1, 3, 3. Wide smoky border on hind wing, as in male, but somewhat paler smoky. Abdomen yellow, when filled with eggs, otherwise whitish. Spiracular dots large, distinct; posterior margins narrowly black. Sternum unmarked. Tails as in male, but the darker joinings may be obscure in the basal portion.

Holotype—Male imago, pinned. Store window at Sheffield, Ala., July 4, 1936 (J. R. T., L. C. Thomsen). No. 1462.1 in Cornell University Collection.

Allotype—Female imago, pinned. Same data. No. 1462.2 in C. U. Collection.

Paratypes—Eight pinned females, two females in alcohol. Same data. No. 1462.3 12 in C. U. Collection.

The somewhat larger size, differences in venation and genitalia, and unusual length of basal fore tarsal joint of male serve to distinguish this species from the allied *S. integrum* McD.

***Stenonema exiguum* Trav.**

A single female imago of this species was found on the radiator of the car on June 27. It had been collected somewhere between Birmingham and Tuscaloosa.

Genus ***Heptagenia*** Walsh

***Heptagenia minerva* McD.**

Hundreds of these tiny and very dainty mayflies crowded the store windows of Sheffield on the evening of July 4. We collected until our cyanide jars would hold no more. Did they come from the Tennessee River, or from some small tributary which we did not see?

***Heptagenia aphrodite* (?) McD.**

A single nymph taken in Cooley Creek on July 4 is doubtfully referred to this species.

Family **Baetidae**

Subfamily **Siphonurinae**

Genus ***Isonychia*** Eaton

***Isonychia fattigi* Trav.**

A single male imago which agrees well with the type material of this species was removed from the radiator of the car on the evening of our arrival in Tuscaloosa, June 27. It had been picked up somewhere between Birmingham and Tuscaloosa. Five female imagos which may be of this species were collected on store windows at Sheffield, July 4.

***Isonychia circe* (?) Trav.**

Nymphs of this species were fairly abundant in the small stream above Spencers Mill, near Tuscaloosa, in Cooley Creek between Birmingham and Tuscaloosa, and in the North River about fifteen miles from the latter city. Several were reared in the Zoology Laboratory.

From nymphs taken in the North River, two females emerged, but no males. Nor were any imagos caught in flight, although two were seen at twilight over North River. The female imago bears a close resemblance to the type material of *circe*, aside from its slightly larger size (body 11 mm.; wing 10 mm.).

Mature nymph (Cooley Creek specimen). Body of female, 11 mm. General color dark red-brown. Basal joints of antenna marked with white; remainder dark brown, no distinct dark band. A wide, pale, median stripe on head is continued backward over the entire thorax, and more indistinctly for the length of the abdomen. Laterally on pronotum, two pale, crescentic marks, and one small round spot. Tibial spine of fore leg relatively long, extending fully one-half the length of the tarsus; curved slightly outward. Spines on outer surface of tibia few in number, 12 to 15 only. Fore leg dark red-brown; pale median and apical bands on femur; claw, and distal half of tarsus pale. On other legs, pale bands also at base of femur and at each end of tibia. Anterior margin of tergite 10 pale, apical portion black. Postero-lateral spine on segment 9 distinctly longer than that on 8. Traces of narrow, black, submedian lines bordering the paler median area, on tergites. Ventrally, traces of pale submedian streaks from anterior margin. Gills largely deep purplish.

A mature male nymph taken at Big Sandy Creek near Cooley may be of this same species. However, the fore femur is largely yellowish; tibial spine shorter and stouter. General appearance similar to Cooley Creek specimens. Body 10 mm., which is rather larger than one would expect the male of this species to be. Other nymphs in the Cornell Collection, from Huntsville, June 18, 1931, agree well with the Cooley Creek specimens except for the fore femur, which again is largely pale.

Subfamily Ephemerellinae

Genus *Ephemerella* Walsh

Ephemerella doris Trav.

One nymph, the only representative of this genus collected in the state, was found in Cooley Creek on July 4.

Subfamily Caeninae

Genus *Caenis* Stephens

Caenis jocosa (?) McD.

Among the swarming multitudes of mayflies on the windows at Sheffield on the evening of July 4, was one female *Caenis*, which appears to be of this species.

Subfamily Baetinae

Genus *Acentrella* Bengtsson*Acentrella propinqua* (?) Wlsh.

One male and one female imago were taken at Sheffield on July 4. As I am not certain that these specimens are *propinqua*, I present a brief description of each sex. Genitalia of male as in fig. 5.

Male imago. Body 4 mm.; wing 4 mm. Turbinate eyes very large, set on very short stalks; bright red-brown. Antennae dusky. Head and thorax black-brown; intersegmental areas of thoracic pleura somewhat paler. Legs white. Wings hyaline. Costa and subcosta dark brown at extreme base; humeral cross vein dusky; longitudinal veins in anterior half of fore wing pale brown, all other veins colorless. Abdominal segments 2-6 white, hyaline; posterior margins very faintly yellowish; at each spiracle, a black circle (*not* a dot). Tergites 7-10 reddish to maroon; sternites paler, somewhat dusky. Tails white, unmarked.

Female imago. Body deep orange; thorax somewhat paler and duller than abdomen. Legs greenish brown. Longitudinal and cross veins, and marginal intercalaries, dark brown.

Genus *Pseudocloeon* Klapalek*Pseudocloeon dubium* (?) Wlsh.

One male subimago and several female imagos, taken on the windows at Sheffield, July 4, are held under this name.

Genus *Centroptilum* Eaton*Centroptilum* sp.

Two female imagos of an undetermined species of this genus were taken at Sheffield, July 4.

FLORIDA

Family EPHEMERIDAE

Subfamily Neoephemerinae

Genus *Oreianthus* Trav.*Oreianthus* sp. No. 1

Mature nymphs of a species of *Oreianthus* much smaller than *O. purpureus* were taken in the Sweetwater Branch, Liberty Co., on Apl. 3, 1927, by Prof. Needham. Since the differences noted between these

nymphs and those of *O. purpureus* might be generic rather than specific (the nymph of the closely-allied genus *Neoephemera* is not known), I designate this species by number only.

Nymph. Body of female, 9–10 mm.; of male, 8–8½ mm. General color, dark red-brown. Head, thorax, and abdomen finely mottled with tiny, pale dots. Crescentic, pale mark on head anterior to median ocellus; diamond-shaped pale spot between base of antenna and inner anterior corner of eye. Lateral margin of pronotum widely flaring at each angle, the middle area concave (see fig. 13). The short, submedian tubercles on anterior margin of this sclerite are closer together than in *purpureus*, almost approximated at the median line. Lateral margin, usually also a narrow, median line, pale yellowish. Projections at anterior margin of mesonotum yellowish, extending out only slightly beyond margin of pronotum. The median tubercle between the wing cases is rather better developed than in *purpureus*. Median spine on mesosternum much as in that species. Legs yellowish to pale red-brown. No distinct markings dorsally. Ventrally, a small, dark dot at apex of trochanter; triangular area on apical portion of femur pale yellowish. Tip of tibia not produced anteriorly, as in *purpureus*. Claws slightly longer and more distinctly curved than in that species.

Median line of abdominal tergites may be wholly pale, or with a pale spot at extreme anterior and posterior margins only. Posterolateral projections paler red-brown than main portions of segments. These projections are rather more flaring than in *purpureus*, especially that on segment 9, which is also relatively longer and more slender. The distinct median tubercle present on the posterior margins of the middle tergites in *purpureus* is here much reduced. Ventrally, a lateral row of dark dots on each side, located at anterior margin of each sternite; traces of a dark streak extending backward from each of these. Ganglionic area of each basal and middle sternite a paler, rounded spot. Tails light red-brown.

GEORGIA

Family EPHEMERIDAE

Subfamily Neoephemerinae

Genus *Oreianthus* Trav.

Oreianthus sp. No. 1

Specimens of this species in the Cornell Collection are from Upatoi Creek, Apl. 29, 1931; Oconee River near Greensboro, Apl. 9, 1931; and Town Creek, May 14, 1931 (P. W. Fattig).

Family **HEPTAGENIIDAE**Genus **Heptagenia** Walsh**Heptagenia thetis** Trav.

Several females, taken at Swamp Creek near Dalton, May 26, 1931 (P. W. Fattig).

SOUTH CAROLINA

Family **EPHEMERIDAE**

Subfamily Ephemerinae

Genus **Hexagenia** Walsh**Hexagenia carolina** Trav.

One male subimago, Clemson College, June 4, 1931 (D. Dunavan).

Hexagenia marilandica Trav.

A male imago and one subimago, Clemson College, June 23, 1933 (B. M. Latham).

Hexagenia kanuga sp. nov. (described under North Carolina)

Florence, May 16, 1929 (O. L. Cartwright).

MARYLAND

Family **EPHEMERIDAE**

Subfamily Potamanthinae

Genus **Potamanthus** Pict.**Potamanthus inaequalis** Ndm.

Imagos of both sexes, taken at Conococheague Park, near Hagerstown, July 22, 1926 (V. Argo).

Potamanthus walkeri Ide

Several nymphs, the Potomac River near Brunswick, June 19, 1926 (V. Argo).

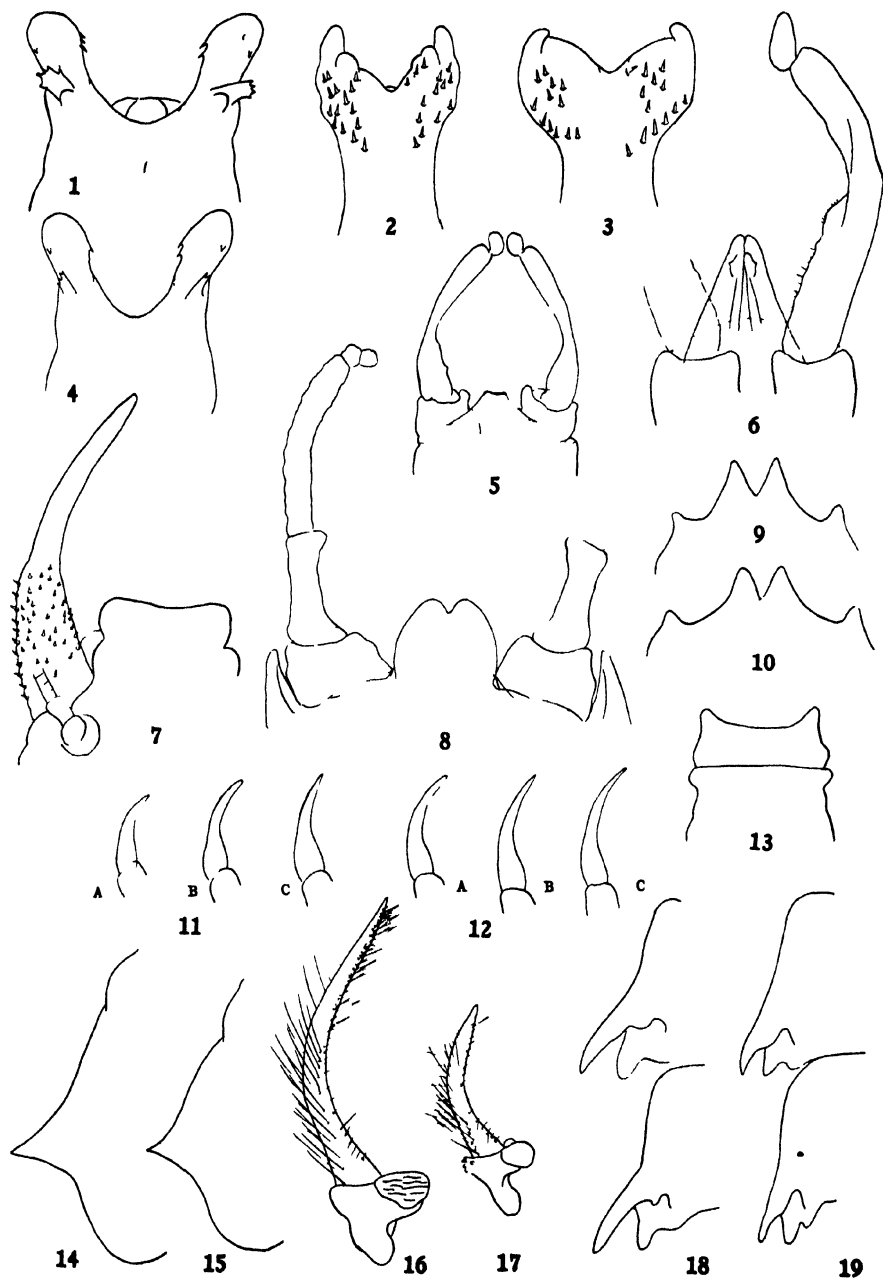
Family **HEPTAGENIIDAE**Genus **Heptagenia** Walsh**Heptagenia lucidipennis** Clem.

About twenty-five specimens, adults of both sexes, were collected from a store window in Frederick, on the evening of May 21, 1936 (J. R. T., R. Rice).

EXPLANATION OF PLATE 6

- Fig. 1. *Rhithrogena rubicunda*. Penes.
Fig. 2. *Ephemerella fratercula* (?). Penes.
Fig. 3. *Ephemerella dorothea*. Variety A. Penes.
Fig. 4. *Rhithrogena fasciata*. Penes.
Fig. 5. *Acentrella propinqua*. Male genitalia.
Fig. 6. *Baetisca thomsenae*. Male genitalia.
Fig. 7. *Potamanthus distinctus*. Mandibular tusk and part of head, nymph.
Fig. 8. *Oreianthus purpureus*. Male genitalia.
Fig. 9. *Baetisca carolina*. Subanal plate, female imago.
Fig. 10. *Baetisca thomsenae*. Subanal plate, female imago.
Fig. 11. *Baetisca thomsenae*. Claws of nymph. A—1st claw; B—2nd claw; C—3rd claw.
Fig. 12. *Baetisca carolina*. Claws of nymph. A—1st claw; B—2nd claw; C—3rd claw.
Fig. 13. *Oreianthus* sp. No. 1. Pronotum and anterior portion of mesonotum, nymph.
Fig. 14. *Baetisca thomsenae*. Lateral portion of mesothoracic shield, nymph.
Fig. 15. *Baetisca carolina*. Lateral portion of mesothoracic shield, nymph.
Fig. 16. *Hexagenia marilandica*. Mandibular tusk, nymph.
Fig. 17. *Hexagenia* sp. Mandibular tusk, nymph.
Fig. 18. *Iron rubidus*. Lateral spines on abdominal segments 6 and 7; nymph.
Fig. 19. *Iron subpallidus*. Lateral spines on abdominal segments 6 and 7; nymph.

PLATE 6



THE FORMATION OF NEW SIPHON OPENINGS IN THE TUNICATE, *STYELA PLICATA**

By W. C. GEORGE

PLATE 7

If one experimentally closes either of the siphon openings of the tunicate, *Styela plicata*, the animal exhibits a remarkable adaptive response. New siphon openings are quickly formed, and the animal resumes normal physiological activity. The directness and effectiveness of this response were such as to indicate a bearing on the mechanistic conception versus a mystical entelechy. An analysis of the processes involved was attempted, therefore, with this point of view in mind.

Styela plicata is one of the larger simple ascidians. Large specimens may measure as much as ten centimeters in length. The inhalent siphon is terminal in position and the exhalent siphon is near it, set only a little way back on the dorsal side. The tunic forming the outer covering of the animal is thick and very tough. The mantle just beneath it is thick and has a highly developed network of muscle fibers circularly, longitudinally, and obliquely disposed. This muscular mantle constitutes the middle layer of the siphon wall. Internal to it, extending as far down as the tentacles, there is a thin layer of tunic continuous over the margins of the mantle at the lips of the siphon with the outer tunic (fig. 1). At the margin of each siphon there are typically four lobes of thickened tunic, and four double lines of pigment extend from the inner lining of the siphon over the margin and a little way down the outside (figs. 4a and b).

In my first experiments I closed either the inhalent or exhalent siphon by sewing, using a large needle and twine. The animals were then

* This investigation was aided by a grant from the Rockefeller Fund for Research in Pure Science at the University of North Carolina. The observations were made at, or upon material obtained at, the United States Fisheries Biological Laboratory, Beaufort, North Carolina. I am indebted to Hon. Frank T. Bell, Commissioner of Fisheries, for permission to use the facilities of the station and to Dr. Herbert F. Prytherch, Director, and other members of the staff for courtesies and assistance.

placed in a live box. A few days later I observed that there was in each case a well developed and normally functioning new orifice a little below the old one. In subsequent experiments, in order to eliminate the possibility of a needle hole being enlarged and used for a new orifice, the original orifices were closed by tying. Animals to be experimented upon were left in some cases in their natural positions attached to the wharf pilings. With these it was necessary to tie the siphons and make the observations at low tide when the animals were exposed. In other cases they were detached and taken into the laboratory, where they were left in water tables or aquarium dishes. A loop of strong twine was carefully placed low around the base of the siphon while the animal was in a relaxed, extended position. The two ends of the looped string would then be quickly and firmly pulled and tied, thus closing the siphon with a strong ligature around its base (fig. 4a). In all several dozen animals were treated in this way. A few animals died from causes other than the ligature and others were lost; but in almost every case a new siphon opening was formed. The new openings might be formed at any point around the siphon just below the ligature.

The time required for the formation of new openings through the siphon wall was in the neighborhood of twenty-four hours, sometimes two or three hours less, sometimes a few hours more. If the ligature was not tightly enough drawn and a very small opening was left, a longer time was taken in the formation of the new orifice; if the ligature was still looser, no new orifice was formed. In those animals in which both siphons were completely closed, the animals died, evidently from lack of aeration; but if a little passage of water was permitted, two new orifices were formed within twenty-four to forty-eight hours.

In order to determine if artificial openings would be used, cuts were made in some animals through the walls of the siphons below the ligature. At the end of eighteen hours these cuts appeared to be healed, and at the end of twenty-four hours other openings formed by the animals themselves were present.

When first formed, the new orifices were merely ragged openings through the siphon walls, but within three or four days the siphons grew out a little distance beyond the ligature and developed specific siphon features, i.e., the typical four lobes of tunic material and four double lines of pigment. The tied-off tip of the original siphon distal to the ligature was left as a mass attached to the side of the siphon (fig. 6).

To understand the mode of formation of the new orifice it is impor-

tant to keep in mind the presence in the mantle of a highly developed and powerful musculature, which extends to the tips of the siphons (figs. 1 and 8), and also the presence of certain reflexes. When stimulated slightly by poking or pinching, an individual will respond by contracting. If the stimulus is strong and persistent the contraction is correspondingly great. In the case of experimental animals the ligature acts as a stimulus to contraction. This stimulus may be supplemented by insufficient food and oxygen resulting from interference with the normal circulation of water. As a result probably of both these stimuli, but especially that of the ligature, the muscle of the mantle contracts strongly, powerfully enough to pull the mantle tissue out of the distal part of the siphon (fig. 3), or if the ligature is too tight to permit this, to tear the mantle in two where the ligature is located. In the latter case one finds distally a ring of mantle left in the tied-off part of the siphon, and proximally the mantle is withdrawn a little distance below the ligature (figs. 2, 5, 7). There is then present a zone just below the ligature where the tunic wall is bare, not reinforced by the muscular mantle. Further contractions of the muscular mantle produce a considerable pressure of water against the inner surface of the tunic below the ligature. In this region there results a softening and thinning of the tunic, which becomes pushed out as a nipple-like projection (figs. 4a and 7). If one examines such an area in a live animal before rupture the tunic is seen to be thin and translucent, whereas it is opaque a little distance away, and in favorable specimens the edge of the mantle may be seen through the translucent tunic of the nipple-like protuberance (fig. 4b). Ultimately a hole is broken through. Observation of an animal with ligated siphon during the hours preceding rupture, reveals periods of forceful contraction followed by periods of relaxation. Very often such an animal will close the unligated siphon and contract forcibly upon the contained water so that a region of the closed siphon where the mantle is torn becomes puffed. Finally the pressure at this point breaks a hole through the unsupported tunic. There is no distinguishable participation of cells in the degeneration, thinning, and rupture of the tunic. Pressure and pressure atrophy appear to be the immediate causes.

During the period in which the mantle of the siphon serves as a nozzle to direct water against a point on the tunic its torn edge is in process of healing. Following rupture the thin tunic lining the inside of the siphon becomes continuous with the outer tunic (fig. 6), the siphon grows longer, and there is ultimately a normal new siphon.

Two or three days or sometimes more are required for complete development of the specific lobes and pigmented lines.

DISCUSSION

Examples of self-adaptation in organisms without highly developed nervous systems are widespread. Every now and then it happens that such organisms are confronted with situations that they may never have met before and with which they have had neither individual or racial experience. Though some perish, others are able to make modifications of structure or reaction that enable them to survive. The observing of apparently purposeful self-adjustment among those animals that survive has given rise to a more or less common belief in the existence of some elemental protoplasmic intelligence, which in some fashion foresees the usefulness of certain structural features and produces them. Some biologists and philosophers support this thesis. Biological literature contains numerous explanations of phenomena apparently based on this belief. Are such teleological explanations valid or is an organism's capacity for self-adaptation to a particular situation inherent in and definitely circumscribed by its anatomical and physiological organization?

On superficial observation the formation of new siphon openings in *Styela plicata* appears to be an example of quick and purposeful self-adjustment. But, as shown above, upon analysis of the events that occur, any apparent mystical entelechy disappears and there is left a result that is a mechanical consequence of particular stimuli acting upon the peculiar structure and reflex organization of the animal.

Analysis of the factors involved in the development of the specific characters that differentiate around the point of rupture is not so easy as is the occurrence of the rupture itself. The fact is that following closure of the original siphon aperture and formation of a new one, certain features that are normally present around the original siphon aperture and which are a part of the heritable structural pattern of the animal develop around the newly formed one. What was originally tunic of the wall of the siphon takes on the somewhat specialized features of the tip after it secondarily and artificially has become tip. Certain relations have changed that may be causal, and the causes may be both organismal and mechanical. After the tip of the mantle comes into relation secondarily with the tunic of the wall of the siphon some organizer in this tip may exert an influence upon the cells of the adjacent tunic, resulting in its further somewhat special differentiation.

That organizers are present in adult tissues seems apparent from the work of Weiss,¹ who found that if the tail of a mature Triton is amputated and the new tissue formed over the wound is transplanted while quite young to the region of the leg, it will develop into a leg; if transplanted later, after the new material has been subjected to the tail determining influences for some time, it will develop into a tail. There is the probability also that the terminal sphincter muscle of the mantle may contribute mechanically through alternate contraction and relaxation towards the development of grooves and ridges in the tunic to which it is adherent.

SUMMARY

Following experimental closure of the inhalent or exhalent siphons of *Styela plicata* new openings are formed below the ligature in about twenty-four hours. There is then developed around these new openings the siphon features characteristic of the species.

Though the end result is quick and very fortunate for the well-being of the animal, no entelechy need be assumed. The formation of the new openings may be explained mechanistically. They appear to result from the stimulus of the ligature acting upon the reflex organization and the peculiar structure of the animal.

The newly developed specific features of the tunic around the opening probably result from the influence of organizers in the mantle acting upon the tunic cells plus the mechanical influence of the constrictor muscles of the siphon.

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UNIVERSITY OF NORTH CAROLINA,
CHAPEL HILL, N. C.

¹ Weiss, Paul 1928 Morphodynamische Feldtheorie und Genetik. Zeitschr. Indukt. Abstam. u. Vererbungsl. Supplement b., 2: 1567-1574.

EXPLANATION OF PLATE 7

All figures are from specimens of the ascidian *Styela plicata*. The drawings are by the author. The photographs are from the Department of Anatomy of Cornell Medical College, New York City. The plate is reduced two-thirds off. Magnifications given are after reduction.

1. Longitudinal section of expanded normal inhalent siphon. The figure shows the thick outer cellulose tunic continuous at the margins or lips of the siphon with a thin layer of tunic which extends down the inner side of the mantle approximately as far as the junction of the siphon with the branchial chamber. Photograph $\times 3\frac{1}{2}$.
2. Longitudinal section of inhalent siphon of experimental specimen Y 1, drawings of which are shown in figures 4a and 4b. The approximate plane of the section is indicated by the straight line in fig. 4b. x — x indicates the position of the ligature, which was removed before sectioning. Observe sections of the ring of mantle torn off from the original siphon and remaining beyond the constriction of the ligature. Photograph $\times 3\frac{1}{2}$.
3. Longitudinal section of inhalent siphon of specimen X 1. As in the specimen shown in figure 2, the mantle has been torn in two where constricted by the ligature. The proximal part of the mantle is withdrawn some distance below the ligature. The specimen was killed during early stages of thinning of the mantle. Photograph $\times 3\frac{1}{2}$.
- 4 a, b. Drawings from different views of specimen Y 1, a section from which is shown in fig. 2. Below the ligature in 4a observe the bulge caused by the pressure of water directed against the bare tunic by the nozzle-like mantle of the siphon. During narcotization and fixation the tension on the tunic was eliminated so that the bulge in fig. 2 is reduced as compared to that in the living animal. In 4b the curved line at * indicates the edge of the torn mantle seen through the translucent tunic. The straight line shows the plane of the section of fig. 2. $\times \frac{1}{2}$.
5. Section of specimen Y 2, which was narcotized and fixed shortly after the tunic had been ruptured at the point where the new lips of the mantle had directed the water pressure. The tunic is very thin in this region. The original distal portion of the mantle is shown in the mass of tunic beyond the constriction caused by the ligature. Photograph $\times 3\frac{1}{2}$.
6. Section of the inhalent siphon of specimen Z 1, which was narcotized and fixed after the formation of a new opening and differentiation of new siphon lips. The tunic of the distal, tied off portion of the original siphon is still present as a mass at one side. Photograph $\times 3\frac{1}{2}$.
7. Hemisection of specimen R 1. The mantle of the original siphon was torn at the ligature, and the distal portion is shown in the mass of tunic beyond the constriction; the proximal portion points towards the bare thinned tunic against which water has been directed. Camera outline drawing $\times 5$.
8. Specimen with new siphon opening seen from the side after removal of the tunic from one side to show the muscular mantle, through which the gonads may be seen. Drawing $\times 1\frac{1}{2}$.

PLATE 7



ANOPHTHALMID BEETLES (FAM. CARABIDAE) FROM TENNESSEE CAVES

By J. MANSON VALENTINE

PLATE 8

INTRODUCTION

Of the seventeen caves visited in Tennessee, ten have yielded new forms of blind carabids of the genus *Pseudanopthalmus*. Out of eleven species and two subspecies found in this state, all except one species (*engelhardti* Barber) have turned out to be new. The following is a list of the caves of Tennessee in which the author has collected; to each productive of anophtthalmids is appended a notation of the fauna insofar as it is known:

- | | |
|--|--|
| 1. Wonder Cave, Monteagle | <i>P. intermedius</i> ; <i>P. humeralis brevis</i> . |
| 2. Crystal Cave, Monteagle | <i>P. humeralis</i> . |
| 3. Johnson's Cave, Monterey | <i>P. robustus</i> . |
| 4. King Solomon's Cave, Cumberland Gap | <i>P. hirsutus</i> . |
| 5. English Cave, Cumberland Gap | <i>P. engelhardti</i> ; <i>P. rotundatus</i> . |
| 6. Lookout Mountain Cave, Chattanooga | |
| 7. Tennessee Cave, Raccoon Mountain | <i>P. digitus</i> ; <i>P. fulleri</i> . |
| 8. Nickajack Cave, Shell Mound | |
| 9. Craighead Cave, Sweetwater | |
| 10. Indian Cave, New Market | |
| 11. Grand Caverns, Byington | <i>P. tennesseensis</i> n. sp. |
| 12. Wash Lee Cave, Livingston | |
| 13. Bunkum Cave, Byrdstown | <i>P. robustus beaklei</i> n. ssp. |
| 14. Piper Cave, Monoville | <i>P. cumberlandus</i> n. sp. |
| 15. Lindsey Williams' Cave, Doweltown | |
| 16. Gin Bluff Cave, Liberty | |
| 17. Dunbar Cave, Clarksville | <i>P. ciliaris</i> n. sp. |

Many promising caves in the state have yet to be investigated and should prove a fruitful field for future research.

For the most part, the above insects have been described in former papers*; four remain to be described herewith.

* See bibliography.

TECHNIQUE

All measurements were made with the aid of a micrometer eye-piece graduated to tenths of millimeters. Curved surfaces were measured as the chords of those surfaces between the desired limits. Since traits of contour and proportion cannot be expressed by linear measurements, an attempt was made to describe such with the aid of indices. Principal ratios were obtained in the following manner:

1. *Head index*: the distance across the eyes (width), measured dorsally from their outer margins, divided by the distance from the outermost extremity of the mandibles in parallel position to the anterior margin of the pronotum when the head is parallel to the latter and normally articulated (length).

2. *Pronotal index*: the greatest transverse distance across the pronotum (width) divided by the distance from the anterior to the posterior pronotal margins measured in the mid-dorsal line (length).

3. *Elytral index*: the greatest transverse distance across the elytra (width) divided by the distance, measured along the suture, from the farthestmost elytral apex to a point directly opposite the apices of the humeral tubercles (the greatest exposed length); when the insect is normally articulated, the latter point coincides exactly with the posterior margin of the pronotum.

4. *Height-length index*: the deepest measurement of the abdomen plus the elytron when normally articulated (height) divided by the *total length*; the latter is the sum of the head, pronotal, and elytral lengths.

5. *Height-width index*: the height divided by the elytral width (*greatest width*).

6. *Antennal segmental index*: the greatest length of the second antennal segment divided by the greatest length of the third, measured dorsally.

7. *Setal index*: the length of the humeral set of four marginal, setigerous papillae divided by the distance between the fourth papilla (most posterior of this series) and the fifth (more anterior of the medial, marginal set of two).

8. *Genital index*: the length of the median lobe of the aedeagus, measured as its chord and including the basal plate, divided by the total length of the insect.

The camera lucida drawings were made by the author.

Pseudanophthalmus ciliaris n. sp.

Holotype ♂, *allotype* ♀: U. S. National Museum. J. M. Valentine, J. C. Beakley, collectors, 1935.

Type locality: Dunbar's Cave, Clarksville, Tennessee.

Holotype ♂ (Plate 8, fig. 1): length 11.7 mm.; width 4.0 mm.; height-length index .23; height-width index .68; head index .56; pronotal index 1.15; elytral index .63; antennal segmental index .82; setal index .30; genital index .18. *Color*: light reddish brown.

Allotype ♀: length 10.9 mm.; width 3.7 mm.; height-length index .23; height-width index .68; head index .58; pronotal index 1.15; elytral index .63; antennal segmental index .80; setal index .34. *Color*: light reddish brown, elytra yellowish brown.

General description: Light reddish brown to pale yellowish brown; elongate, convex; dorsal surface reticulo-alutaceous. *Antennae*: long. *Head*: narrow, rather elongate. *Pronotum*: very sparsely and finely pubescent; narrow, sides gently sigmoid; posterior angles acuminate; lateral tubercles of the posterior margin dentate, produced. *Elytra*: narrow; convex, basal disc depressed, humeri rounded; margins narrow; striae distinct, impunctate, apical striae arcuate; intervals convex, each with a narrow band of from one to four rows of fine, rather long, straight pubescence. *Legs*: long. *Aedeagus* (Plate 8, fig. 1a): large; median lobe straight distal to basal bulb which is abruptly deflected, uniformly tubular; apex slightly dilated and not produced in sagittal view, median lobe slightly constricted over apical third in lateral view; lateral pieces attenuate, bearing four, long setae.

This species was taken on April ninth in considerable numbers on the mud banks of the principal cave stream and on rotting boards and muddy floors in passageways and stalactitic chambers. The author has never seen an anophthalmid more abundant or more ubiquitous. A type series of forty were taken but only a fraction of those seen were captured. It is surprising to find a population of this size in a cave which has been commercialized; perhaps the presence of a sizable cave stream and the extensiveness of the cavern will insure the safety of the colony as has been the case in the Mammoth Cave complex.

Ciliaris resembles *pubescens* (Horn) of Cave City, Kentucky, in form and pubescence but is smaller, more elongate, and less hairy. These two species are representatives of colonies from two separate drainage

systems, the former being located on the Red River, a tributary of the Cumberland, while the latter belongs to the Green River system. They might have originated from a parent stock dwelling in that region of Kentucky which divides these two water-sheds; however, a comparative study of genitalia reveals that *ciliaris* and *pubescens* are now widely divergent.

***Pseudanophthalmus cumberlandus* n. sp.**

Holotype ♂, *allotype* ♀: U. S. National Museum; J. M. Valentine, J. C. Beakley, collectors, 1935.

Type Locality: Piper Cave, Monoville, Tennessee.

Holotype ♂ (Plate 8, fig. 2): length 9.5 mm.; width 3.4 mm.; height-length index .22; height-width index .62; head index .64; pronotal index 1.17; elytral index .65; antennal segmental index .80; setal index .76; genital index .14. *Color*: pale yellowish brown, slightly reddish.

Allotype ♀: length 9.5 mm.; width 3.4 mm.; height-length index .22; height-width index .62; head index .62; pronotal index 1.15; elytral index .64; antennal segmental index .80; setal index .70. *Color*: pale yellowish brown.

General description: pale yellowish brown; transverse, rather flat; finely reticulo-alutaceous, more distinctly and regularly so on head. *Antennae*: normal. *Head*: narrow, not elongate. *Pronotum*: normal, sides gently sigmoid; posterior angles rectangular, rounded; lateral tubercles of posterior margin distinct, obtuse; disc bearing several rather long setae. *Elytra*: transverse, rather flat, basal disc not depressed, humeri angulate; margins rather wide; striae distinct, impunctate, apical striae arcuate; intervals feebly convex, glabrous. *Legs*: normal. *Aedeagus* (Plate 8, fig. 2a): small; median lobe short, arcuate without distinct basal flexure, tapering over the distal third to a rounded apex; transfer apparatus long, sigmoid in lateral view; lateral pieces armed with four setae.

Five specimens in all were taken on the muddy floor of a moist, lateral passage, April seventh. The cave is a spacious one and is said to extend as a simple passage two miles to the Cumberland River. For the most part it is dry, there being no cave stream. It is situated at Monoville, four miles north of Carthage on the north side of the river.

In form of body and general plan of aedeagus, *cumberlandus* is remini-

scent of *horni* from the rock quarries of Lexington, Kentucky. However, the resemblance must be considered one due to parallelism rather than relationship until caves of the water-shed shared by the Kentucky, Green, and Cumberland Rivers yield a common ancestor whose derivatives may have wandered to the respective points, a hundred and fifty miles apart, where the two species now occur. *Cumberlandus* differs noticeably from *horni* in being considerably larger and more elongate and in having a far greater genital index and a much smaller transfer apparatus.

***Pseudanopthalmus robustus beaklei* n. ssp.**

Holotype ♂, *allotype* ♀: U. S. National Museum; J. M. Valentine, J. C. Beakley, collectors, 1935.

Type locality: Bunkum Cave, Byrdstown, Tennessee.

Holotype ♂ (Plate 8, fig. 3): length 9.5 mm.; width 3.4 mm.; height-length index .22; height-width index .62; head index .60; pronotal index 1.20; elytral index .67; antennal segmental index .76; setal index .69; genital index .20. *Color*: light reddish brown, elytra paler.

Allotype ♀: length 10.6 mm.; width 3.7 mm.; height-length index .22; height-width index .62; head index .58; pronotal index 1.21; elytral index .65; antennal segmental index .76; setal index .65. *Color*: light reddish brown.

General description: light to pale reddish brown, rather transverse, moderately convex; very finely reticulo-alutaceous, more distinctly and regularly so on head. *Antennae*: normal. *Head*: normal. *Pronotum*: transverse, quadrate; sides gently arcuate with rather wide margins; hind angles rectangular, sharp; basal tubercles dentate, produced; disc glabrous. *Elytra*: transverse, convex, with sutural and basal regions flattened; humeri full, rounded; margins wide, distinctly serrate at humeri; striae deep, regularly, deeply but sparingly punctured, apical striae arcuate; intervals convex, glabrous. *Legs*: normal. *Aedeagus* (Plate 8, fig. 3a): long; median lobe evenly, narrowly cylindrical, slightly arcuate, constricted over distal fourth, abruptly bent at basal flexure; apex spatulate-rounded in sagittal view, elongate-knobbed in lateral view; lateral pieces relatively short, arcuate, bearing four setae, the central pair of which is long.

Robustus beaklei resembles true *robustus* in nearly every respect; the punctuation of the elytral striae is slightly less distinct and regular and

the head is a little broader in the typical form. The main point of difference lies in the aedeagus which is straighter and narrower in *beaklei* and possesses much shorter and more curved lateral pieces.

It is remarkable that two such closely related forms as *robustus* and its vague subspecies should occur so far apart geographically. Fully forty miles intervene between the two type localities. Both caves, however, drain into the Cumberland River to the east of which Monterey (type locality of *robustus*) and Byrdstown are located.

The author takes pleasure in naming this anophthalmid in honor of Mr. John C. Beakley who first discovered the insect in Bunkum Cave and who aided in collecting the paratype series of twenty in the author's collection. They were taken running over the banks of the cave stream which nearly filled the narrow passageway throughout the quarter mile or more of easily traversed cavern. The cave water flowed out through the mouth of the cave, continuing as a surface stream, a condition seldom encountered in caves supporting an anophthalmid population. The date of collecting was April sixth.

***Pseudanophthalmus tenesensis* n. sp.**

Holotype ♂: U. S. National Museum; J. M. Valentine, collector, 1934.

Allotype ♀: col. Valentine; J. M. Valentine, collector, 1932.

Type locality: "Grand Caverns", Byington, Tennessee.

Holotype ♂ (Plate 8, fig. 4): length 8.00 mm.; width 2.65 mm.; height-length index .20; height-width index .60; head index .61; pronotal index 1.15; elytral index .62; antennal segmental index .81; setal index .61; genital index .15. *Color*: pale yellowish brown, head and pronotum slightly darker and redder.

Allotype ♀: length 7.8 mm.; width 2.5 mm.; height-length index .19; height-width index .60; head index .64; pronotal index 1.15; elytral index .59; antennal segmental index .81; setal index .67. *Color*: very pale yellowish brown, head slightly darker.

General description: pale yellowish brown; elongate, flattened dorsally; very finely reticulo-alutaceous, more regularly and coarsely so on head. *Antennae*: normal. *Head*: narrow. *Pronotum*: narrow, sides nearly straight over basal two thirds, very gently arcuate; lateral margins narrow; posterior angles rectangular, sharp; lateral tubercles of the basal margin dentate, not produced. *Elytra*: narrow, disc flat, humeri rounded, not well developed; margins narrow; striae almost obliterated, apical striae sinuate; intervals feebly convex, bearing rather long, sparse pubescence. *Legs*: normal.

Aedeagus (Plate 8, fig. 4a): average size; median lobe strongly arcuate in lateral view, elevated in center, tapered into a truncate apex over distal half; in sagittal view, nearly straight, apex tongue-shaped; basal bulb not well differentiated, basal flexure very obtuse; transfer apparatus long, internal piece sinuate; lateral pieces armed with three, short setae.

Judging particularly by its genitalic equipment, *tenesensis* is related most closely to three other species occurring to the west and north and near the source of the Tennessee river: *humeralis* from Monteagle, Tennessee, *lodingi* from Huntsville, Alabama, and *engelhardti* from the Powell River at Cumberland Gap. To this group *tenesensis* unquestionably belongs and forms another link in the chain of related yet isolated species found from extreme northeastern Tennessee to northeastern Alabama.

A female was taken on March 15th, 1932, and a male on May 24th, 1934, running over wet, rotting boards. "Grand Caverns" is a small, rather dry cave without a cave stream. In spite of its habitat being commercially exploited, the small colony of this minute species is probably holding its own.

REMARKS

In drawing conclusions regarding the effect of cave life on anophthalmids and the consequent isolation which is undoubtedly conducive to the evolution of species, a striking example of the independability of this factor must be borne in mind. At Monteagle, Tennessee, Crystal Cave and Wonder Cave, only a thousand feet or so apart, yield two distinct forms of a single species, namely: *humeralis* and *humeralis brevis*. These can be distinguished externally by the more angular and produced humeri characteristic of the colony from Crystal Cave (*humeralis*). This difference, of course, may be interpreted as evidence that the two colonies do not and probably cannot interbreed. However, *lodingi*, as it occurs in Shelta Cave, Huntsville, Alabama, has been found to be identical in external as well as internal features with a colony living at the bottom of Natural Well, 320 feet below the surface of Monte Sano and several hundred feet above Shelta Cave, some ten miles distant (collected by Drs. Walter Jones and H. P. Loding); it is likewise identical with another colony, discovered by the same collectors, thirty miles to the northeast at Aladdin Cave, Madison Co.

LITERATURE CITED

JEANNEL, R.

1931. Révision des Tréchinæ de l'Amerique du Nord. Archiv. de Zool. Exp. 71: 403-499.

VALENTINE, J. M.

1931. New Cavernicole Carabidae of the subfamily Trechinae Jeannel. Jour. Elisha Mitchell Sc. Soc. 46: 247-258.
1932. A classification of the genus *Pseudanophthalmus* Jeannel with descriptions of new species and notes on distribution. Jour. Elisha Mitchell Sc. Soc. 47: 261-280.

Errata in the above paper:

Page 267: third line from top, for *punctatus* read *gracilis*.

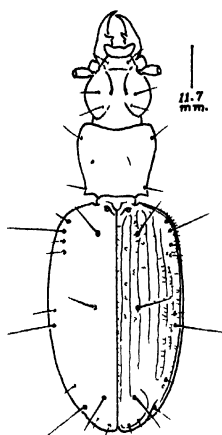
Page 274: seventeenth line from bottom, for Antennal index 4.5 read Antennal index .70.

EXPLANATION OF PLATE 8

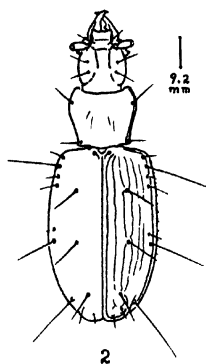
The measured line indicates actual size of object figured.

- Fig. 1. *Pseudanophthalmus ciliaris* n. sp., holotype ♂. Dunbar Cave, Clarksville, Tennessee.
Fig. 1a. Aedeagus of same, sagittal (lower) and lateral (upper) views.
Fig. 2. *Pseudanophthalmus cumberlandus* n. sp., paratype ♂. Piper Cave, Monoville, Tennessee.
Fig. 2a. Aedeagus of same, sagittal (lower) and lateral (upper) views.
Fig. 3. *Pseudanophthalmus robustus beaklei* n. ssp., holotype ♂. Bunkum Cave, Byrdstown, Tennessee.
Fig. 3a. Aedeagus of same, sagittal (lower) and lateral (upper) views.
Fig. 4. *Pseudanophthalmus tenesensis* n. sp., holotype ♂. "Grand Caverns", Byrdstown, Tennessee.
Fig. 4a. Aedeagus of same, sagittal (lower) and lateral (upper) views.

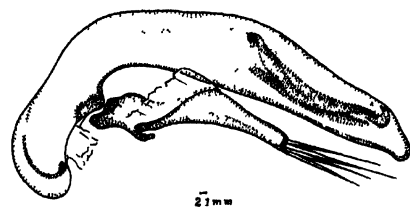
PLATE 8



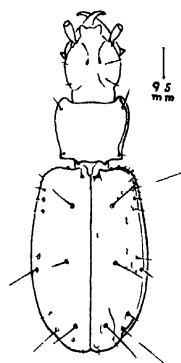
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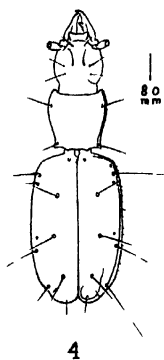
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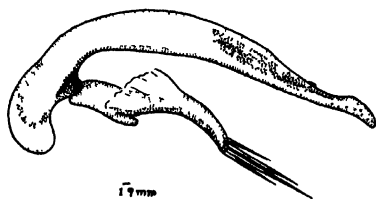
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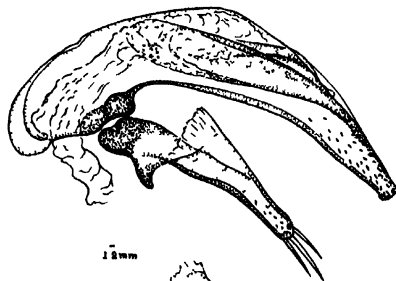
4



2a



3a



4a

A CYTOLOGICAL STUDY OF THE SKIN GLANDS OF THE DUSKY SALAMANDER

By MARY IDOLENE McMANUS

PLATES 9-11

INTRODUCTION

Skin glands of Amphibians have, since 1840, been favorite objects of study. In spite of the great numbers of investigations carried out, there is still no general agreement concerning many of the phases of the work, such as (1) origin of glands, (2) number and kinds of glands, (3) origin and character of secretion, (4) gland musculature, (5) nature of the regeneration process.

The present work was undertaken with the idea of studying anew some of these unsettled questions; it deals especially with kinds of glands, differentiation and regeneration phenomena. Observations on gland musculature are also reported.

The work was suggested by and done under the direction of Dr. Geo. T. Hargitt, to whom the writer is deeply indebted for valuable aid and criticisms.

MATERIALS AND METHODS

Specimens of dusky salamander, *Desmognathus fuscus* (Rafinesque) were taken from the same general locality in the Duke Forest, in January, February, October, November, December 1934, January, February and March 1935. Most material was fixed within a few hours after collection, though some specimens were kept in the laboratory, under conditions similar to those of the natural environment, from one to forty days for study of regeneration of glands.

Of the common fixing fluids employed, Zenker's with formalin instead of acetic acid was by far the most satisfactory for the skin in its entirety. For general histological study, Ehrlich's hematoxylin followed by eosin gave good results. As special differentiating stains Hoyer's thionin and Mallory's triple connective tissue stain were extensively employed with excellent results. Methylene blue and Mayer's mucicarmine and mucihaematin were also used occasionally.

TYPES AND DIFFERENTIATION OF SKIN GLANDS OF AMPHIBIA

Size, character of secretions, and histological structure are criteria that have been used to differentiate types of skin glands of Amphibia. Size alone was the basis used by earliest investigators. Engelman ('72) used the terms 'Schleimdrüsen' and 'Körnerdrüsen', and these terms have since been generally employed. The majority of investigators believe that the skin glands of Amphibia are of two distinct types, mucous and granular, but some few investigators have found only mucous glands. Others hold that only one type, the granular, is present as a mature form. A mixed type is observed by many investigators, but various interpretations are given to it. Nicoglu ('93) in Triton regards the mixed gland as a temporary condition, caused by replacement of a granular gland by a mucous one or vice versa; Nirenstein ('08) observes temporary mixed glands in *Salamandra maculosa*, arising as mucous glands and replaced by granular ones. Hoyer ('90) describes a mixed condition in some poison glands and suggests the possibility of transformation of mucous cells into poison cells, but direct proof is not given. Esterly ('04) is of the opinion that the mucous sac, observed in the poison glands of Plethodon, enlarges and metamorphoses into a poison gland, and mucous cells transform into poison cells. However, he was not able to demonstrate such a transformation. Nirenstein also by indirect proof concludes that such a transformation takes place in cells of *Salamandra maculosa*. Dawson ('20) thinks the mixed condition as observed in Necturus is probably permanent. Theis ('32) finds no evidence of transformation of mucous glands into poison glands or vice versa.

In *Desmognathus* I find that skin glands are functionally mucous and granular, as shown by differential stains (figs. 1, 2). Several types of mixed glands are present, resulting from transformation of mucous cells into granular cells; or from regenerating processes. Evidence presented by my preparations makes it possible to demonstrate that mucous cells actually transform into granular cells. In glands of *Desmognathus*, stained in thionin, cell after cell is noticed in which the basal portion contains a homogeneous purple mucous secretion, and the free end of the same cell contains a blue mass of distinct granules. Mallory's stain is even more convincing; gland after gland is observed in which some cells show a true mucous reaction; a neighboring cell of the same gland contains granules which stain blue, exactly as the first cell; other granules in the same cell are greenish, others yellow-orange, and red (figs. 6-8, 13, 15-17). Mallory's stain is known

to stain mucus blue, and other investigators using it state that it stains granules red, thus the above evidence is proof that transformation of a mucous cell into a granular one takes place. Nirenstein holds that direct proof would consist in finding, in the same cell, some granules giving the mucous reaction and others giving the typical granular reaction. This condition occurs abundantly in my preparations. In addition to this type, other types of mixed glands are observed, involving the old gland and a new anlage which is associated with it. In practically all completely matured granular glands there is noticed the structure designated as the 'Heidenhain gland sac.' This is present in all degrees of development from a few indifferent cells in the intercalary region to an almost completed granular gland (figs. 13-17). In longitudinal sections of glands this sac, in its early stages, is observed as a very inconspicuous, non-differentiated cell mass, near the opening of the duct in the intercalary region; as development proceeds, the cells increase in size and number, and most commonly begin to grow around the old gland wall between the muscle layer and old poison cells. At first the structure consists of a solid mass, then with the acquisition of a lumen, sections show two layers of cells present, one on each side of the developing cavity. Typically the cells of the inner layer are smaller than those of the outer layer, and in many cases are seen to consist of only a very fine protoplasmic strand with or without nuclei. This layer may consist of low epithelial cells in which no granules can be observed, or more commonly of cells filled with granules like those in cells in the outer layer (figs. 14, 15, 17). As development proceeds some cells are observed in both layers in which the entire contents give the mucous reaction, other neighboring cells react like poison granules, and still others give reactions intermediate between these, a condition that may also be found in the old gland itself (figs. 13-17). In such anlagen the red granules are usually nearest the free end of the cells, followed by yellow-orange ones, with blue ones nearest the base (figs. 15-17). Similar results are obtained with thionin, for some cells of the bud stain a homogeneous purplish color, while others have deep blue granules at the free end of the cells with the homogeneous purple near the base. Other adjoining cells are completely filled with blue granules. The number of differentiated cells forming the bud varies from five or six to many; and quite often the bud is seen to extend as far as the posterior pole of the old gland, and in some cases it occupies two thirds or more of the gland globus (figs. 14-16). Occasionally the bud has developed so far in size and form as to appear

like a mature mucous gland, with epithelium low and cubical, and the majority of cells containing various colored granules, while a smaller granular mass, remains of the mother gland, is pushed far to one side of the large bud, which has almost completely filled the old gland cavity. All basal cells of the bud are differentiated, but those nearest the upper pole still take the cytoplasmic stains. Cells of the outer layer of the bud appear to grow faster than those of the inner layer, and always react to differential stains, while many times those of the inner layer are still undifferentiated and take only protoplasmic stains (fig. 17). Another similar mixed gland is observed in a few glands of *Desmognathus*, except in this case the type of gland involved is the mucous one (figs. 11, 12). Ingrowth is the same for this type as that described for the granular one; the ingrowing bud is evidently pushing away the old mucous epithelium and taking its place (figs. 11, 12). As the bud increases in size the mucous part becomes smaller, but retains its mucous character, as shown by its color reaction to specific stains. The first two conditions are typical, occurring in great abundance, while the last is met with only occasionally.

General gland structure as observed in *Desmognathus* does not differ essentially from that found in other *Amphibia* (figs. 1, 2). The muscles however, need some comment. Observers generally agree that there is present a muscular layer for granular glands, but very few report it for mucous glands. Gland muscles in a few cases have been reported to extend into the epidermis, and to connect by intercellular bridges to epidermal cells. In *Desmognathus* well developed muscle fibers are present on both mucous and granular glands (figs. 1, 2). In longitudinal sections of glands, fibers are much more conspicuous and numerous in the intercalary region, but they are not restricted to this region alone. The muscularis stains a purplish color in Mallory's stain and is easily distinguished from the dense blue connective tissue with which it is in close contact (figs. 6, 14, 17). At the anterior pole of glands broad bands of muscles insert on *Schaltzellen* (Nicoglu '93), and pass over the outer surface to surround them before passing into the epidermis (figs. 1, 2, 5). Dilator and constrictor muscles can be observed around mouths of ducts, just within the epidermis, above the outer dense layer of connective tissue of the corium (figs. 3, 4). Esterly is the only other investigator who has reported such muscles, and so far as I can determine these in *Desmognathus* are the same in structure and function as those in *Pléthodon* (Esterly '04).

All types of glands develop from the epidermis, and are in the earliest

stages non-differentiated. The first signs of differentiation can be observed before a lumen forms, in that cytoplasm in some cells begins to take on characteristic colors of mucus with differential stains. Differentiation proceeds in two directions: the cells either become arranged to form a low cubic epithelium, characteristic of typical mucous glands, or they become tall and cylindrical to form giant epithelial cells of granular glands. Such changes as the following are characteristic: Glands without a lumen, cells of which give mucous reactions; similar glands in which the majority of cells show the typical mucous reaction with a few cells entirely granular, while still other cells are mixed. Thus a transformation may be followed from mucoid granules through a mixed series to final forms. This same process takes place in regenerating gland anlagen (figs. 13-17). Some large Leydig cells of the old gland, in which cell walls are still distinct, show mucoid granules and mature granules existing side by side (fig. 13). The Heidenhain gland sac is present in some glands thus characterized, and shows the same conditions as given for giant cells of the old glands (fig. 13). Glands can be observed in which all cells contain only mature granules, but there is an indication of mucous material intermingled in the cell contents. Finally, glands show only mature granules in enormous cells completely filling the gland cavities. Cell walls eventually rupture, and the granular mass is free in the gland globus.

My preparations thus show no evidence that the types of glands in very early stages of development are mucous and granular. Origin and development are the same up to a certain point when differentiation takes place. Gland anlagen are at first indifferent and later become mucous. These differentiate in two directions to become either functional mucous glands, so prevalent in the skin, or large granular glands, so characteristic of the skin of most Amphibia. Thus I agree with Nirenstein that poison glands may arise from mucous glands. In my material, evidence is furnished in a limited number of cases to show that a typical mucous epithelium is replaced by a poisonous (granular) one in the way described by Nirenstein—granular epithelium grows from the intercalary region between mucous cells and gland muscles to push mucous epithelium from its original position, and to take its place (figs. 11, 12). My observations support the following conclusions: Granular glands are formed typically by metamorphosis of a young non-functioning mucous gland into a granular one; less often by metamorphosis, cell by cell of a typical matured mucous gland; quite commonly by regeneration of the old poison gland by a new gland

bud growing within the old poison cavity; and occasionally by a new gland bud growing into and replacing old mucous epithelium of a former gland (figs. 6-8, 11-17). Since mucous glands are differentiated first, and since granular glands are formed from mucous glands, there is logic in Nirenstein's belief that mucous glands are phylogenetically the older glands.

Although the most recent investigators of Amphibian skin glands, Frederikse ('31) and Theis ('32), find a separate origin and development of mucous and poison glands, and hold that there is no genetic relation between the two, mucous glands being always only mucous glands and poison glands only poison glands, evidence gained by the use of Mallory's stain and Hoyer's thionin furnishes conclusive proof that in *Desmognathus* mucous glands metamorphose into granular glands during their histogenesis (figs. 6-8).

FATE AND REGENERATION

It is the consensus of opinion that cells of mature poison glands pass bodily into the secretion mass. The emptied gland cavity then degenerates completely, and a new gland regenerates to take the place of the depleted one. Depleted granular glands are replaced by either of two definite processes—by entirely new origin (Engelman '72, Junius '96, Muhse '09) or by a regeneration process involving a gland bud (Heidenhain gland bud) which grows into and fills the old gland cavity (Nicoglu '93, Heidenhain '93a, Vollmer '93, Esterly '04). My observations support the latter.

Nirenstein ('08), as previously stated, observed an epithelial sac in the glands of Triton and *Salamandra maculosa*, which he says is identical with the Heidenhain gland sac, but which he interprets as a method by which a mucous gland is replaced by a granular one, rather than a regeneration process. Heidenhain and Nicoglu report that the gland sac has the character of young poison cells (granular), but Nirenstein says that without exception the sac is mucous in character. Bristol and Bartelmez ('08) and Shipley and Wislocki ('15) find that the poison gland degenerates, the remains are removed, and one of the five or six undeveloped glands found about the mouth of the functional gland grows down to occupy the place of the former gland. Frederikse ('31) concludes that glands of frog skin can be made to degenerate by electrical stimulation and by injection of adrenalin. The degenerated glands then regenerate themselves by a transformation of the so-called covering cells (smooth muscle cells).

My preparations show that the fate and regeneration of granular glands are primarily the same as described by Nicoglu ('93) for Triton; death and degeneration follow the formation and expulsion of the secretion. The regenerating gland anlage originates similarly, and the method of growth is likewise similar. However, in one essential point I disagree with Nicoglu, Heidenhain, and Vollmer. They state that the gland bud is granular. Esterly maintains that it is always mucous in character. I am forced to take a stand that is not in accord with either, for in *Desmognathus* this bud certainly is not granular to begin with, nor does it remain mucous for any length of time (figs. 13-17). All of Esterly's figures show it as a pure mucous bud regardless of its size, but he suggests that it takes over the function of the replaced granular gland. My material shows that a small bud, consisting of only three or four cells, may be observed occasionally as entirely mucous, but in general the gland bud is either mixed or pure granular. The mixed condition is most abundant, in which some cells are mucous, other cells are mucous and granular, and others are only granular (figs. 13-17). This means that mucous cells are metamorphosing into granular cells, and since practically every gland bud shows this condition rather than pure mucous or pure granular characteristics, I hold that the cells of the bud are first differentiated as mucous cells which one by one quickly change to granular cells, just as is the case with cells of the mother glands. Since the metamorphosis is not simultaneous for the cells, a temporary mixed bud results, and granular glands as well as granular cells go through a mucous or mucoid-like stage before reaching maturity.

Nirenstein ('08) opposes the view of Nicoglu, Heidenhain, Vollmer, and Esterly in regard to the gland sac. It results from a process of replacement of mucous epithelium by granular epithelium, as granular epithelium grows in between mucous epithelium and muscles of the gland. As fast as the number of granular cells increases with development, the number of mucous cells decreases. My preparations show this can not be the case generally, because the fewer the number of cells present in the gland bud the more mucous it is, and, as the bud increases in size and number of cells, the new granular cells become more numerous and the mucous less numerous, while at the same time the old granular mass gradually becomes smaller (figs. 13, 15, 16). This could not be the case if the mucous epithelium were being replaced by granular epithelium in the way described by Nirenstein. Other evidence against this view is that new non-differentiated cells of the

bud are seen growing down from the intercalary cells which gradually metamorphose into mucous cells before becoming granular ones (figs. 13-16). In view of the evidence listed I repeat that mucous cells metamorphose into granular cells, and that degenerating granular glands are regenerated by the ingrowth of a new gland anlage into the old gland cavity. On the other hand some evidence is presented by my material to show that Nirenstein was right in what he saw and in his interpretation of such. Glands are occasionally present in *Desmognathus* in which the same type of gland bud occurs, and grows downward in the old gland cavity in the same way, but as it grows it pushes away mucous epithelium from the old gland wall instead of granular epithelium; there is accordingly formed a mucous bud which gradually becomes smaller as the granular mass increases in size just as is described by Nirenstein (figs. 11, 12). In the first case an old granular gland is involved, while in the second an old mucous one; both types are present in one slide and thus it is obvious that both authors can be right in their observations and interpretations.

Few investigators have seriously attempted to solve the fate of mucous glands and since practically no evidence is available to show the fate and regeneration of these, it seems worth while to examine the possibilities suggested by Arnold ('05). No evidence is gained in my study to support the possibility that new epithelium may be developed by a downgrowth from the epithelium into the old gland. The possibility that the cells may not be destroyed in secretion, but rejuvenate themselves from the non-metamorphosed part of the cytoplasm seems plausible. Occasionally my preparations show three or four basal cells in the mucous glands as low, cubical epithelium in contrast to the much larger mucous cells in other regions of the gland. As a rule the basal cells have begun discharging their mucus before the other cells of the gland, as is shown by the free secretion in the gland lumen. Whether the smaller size of these basal cells is due to the fact that they are being used up in secretion, or whether it is new epithelium which has grown from a small area of non-metamorphosed protoplasm at the base of the cells, I can not definitely say. The other possibility suggested by Arnold, that some cells may be destroyed in secretion and their loss offset by the differentiation of cells from the intercalary region, is a reality in *Desmognathus* (figs. 9, 10). A definite method of regeneration involving new cells growing from the intercalary region is certain. The cells budded from those of this region are observed to grow downward around the old gland cavity in a layer from the place

of origin. Varying degrees or development of this layer are observed, but no cases are noted in which this new growth has reached further than about half the distance of the gland body. Where this method of renewal is observed, the old mucous cells in the region of the new cells are completely degenerated; no cell walls or nuclei are apparent, and only a mass of secretion is in the place where former mucous cells once existed (figs. 9-10). Quite often these new cells show slight differentiation at their free ends into new mucous epithelium, and often the cell is entirely filled with secretion, but the epithelium is low and smaller in size than the old cells of the glands (fig. 10). It is known to be new epithelium and secretion because of the small size and of the purplish tinge intermingled with the blue of the typical mucous reaction to Mallory's stain.

Arnold suggests that the time of year may have some influence on the regenerative processes in the gland. In my materials no preparations made in the months of September, October and November show any indication of the regenerative process just described for the mucous glands, but in preparations made from December to March this method of regeneration is observable, both in specimens just captured and in specimens kept in the laboratory from one to six weeks. Practically every matured mucous gland in these preparations shows regeneration in some degree; those specimens kept in the laboratory show the regenerating process to a greater degree, and the ones killed after a week's stay show the process less advanced than those of two weeks' or longer. I think perhaps this is due to the fact that when these specimens were captured they were stimulated to such an extent that more mucus was given off than is done in the general routine of the animal's life, thus necessitating destruction of cells in the sudden response to meet new demands. As a result, more abundant and sudden regeneration is required, and the longer the length of time allowed (as cited) after such an experience, the greater the degree of regeneration. However this may be, regeneration is unmistakably taking place in mucous glands.

SUMMARY

Mature functional glands of *Desmognathus* are of the mucous and granular types. Mixed glands are abundant. All types possess a muscularis around the gland globus, and special triangular shaped dilator and constrictor muscles are found around the mouths of gland ducts, just above the corium.

Glands are of only one kind in early development. No differentia-

tion is evident in the earliest stages, since contents of glands at this time give neither the mucous nor granular reactions characteristic of mature glands. Differentiation of primal glands into typical mature mucous glands and typical granular glands takes place. Granular cells and glands pass through a mucoid stage during histogenesis; some contain both mucous secretion and granular secretion at the same time, and intermediate stages can be traced from mucous cells to granular ones.

Granular glands are destroyed in the elaboration and expulsion of secretion, and are regenerated by cells growing from the intercalary region. A new gland bud is thus formed which eventually fills the cavity of the old gland. Cells of the regenerating bud are first mucous and later granular; one cell after another gradually changes from the mucous type to the granular type; thus both young and parent gland pass through a mucous stage in development.

Mature granular glands commonly arise from indifferent glands growing from the epidermis; these pass through a mucous-like stage before reaching their final form. Less commonly they form from matured functioning glands by transformation of mucous epithelium into a granular one, or by a regeneration of an old granular gland by means of an indifferent gland bud transforming into a granular one. Occasionally they arise by replacing an old worn-out mucous gland as a new bud grows into its cavity, or by a regeneration of the old granular gland by a similar gland bud.

Mucous glands function more or less continuously for a varied period of time. In case mucous epithelium deteriorates or is destroyed it may be regenerated by new indifferent cells growing from the intercalary region into the old gland cavity.

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LITERATURE

ARNOLD, J.

1905. Über Bau und Sekretion der Drüsen der Froschhaut; zugleich ein Beitrag zur Plasmosomen-Granulalehre. Arch. mikr. Anat. 65: 649-666.

BRISTOL, C. L., AND G. W. BARTELMER

1908. The poison glands of *Bufo agas*. Science, N. S. 27: 455.

DAWSON, A. B.

1920. The integument of *Necturus maculosus*. Jour. Morph. 24: 467-589.

- ENGELMANN, T. W.
1872. Die Hautdrüsen des Frosches. Eine physiologische Studie. Arch. ges. Physiol. 6: 97-158.
- ESTERLY, C. C.
1904. The structure and regeneration of the poison glands of *Plethodon*. Univ. California Pub., Zool., 1: 227-268.
- FREDERIKSE, A. M.
1931. Die Regeneration der Hautdrüsen des Frosches. Zeitschr. mikr.-anat. Forsch. 24: 441-455.
- HEIDENHAIN, M.
1893a. Die Hautdrüsen der Amphibien. Sitzb. phys.-med. Gesell., Würzburg, Jahrg, 1893, S. 52-64.
- HOYER, H.
1890. Über dem Nachweis des Mucins in Geweben mittels der Färbemethode. Arch. mikr. Anat. 36: 310-375.
- MUHSE, E. F. F.
1909. The cutaneous glands of the common toads. Am. Jour. Anat. 9: 321-361.
- NICOGLU, P.
1893. Über die Hautdrüsen der Amphibien. Zeitschr. wiss. Zool. 56: 409-487.
- NIRENSTEIN, E.
1908. Über den Ursprung und die Entwicklung der Giftdrüsen von *Salamandra maculosa* nebst einem Beitrage zur Morphologie des Sekretes. Arch. mikr. Anat. 72: 47-141.
- SHIPLEY, P. G., AND G. B. WISLOCKI
1915. The histology of the poison glands of *Bufo agui* and its bearing upon the formation of epinephrin within the glands. Contr. Embr., Carnegie Inst. Wash. 3: 71-90.
- THEIS, A.
1932. Histologische Untersuchungen über die Epidermis im Individual-cyclus von *Salamandra maculosa* Laur. Zeitschr. wiss. Zool. 140: 356-420.
- VOLLMER, E.
1893. Ein Beitrag zur Lehre von den Regeneration, speziell der Hautdrüsen der Amphibien. Arch. mikr. Anat. 42: 405-423.

EXPLANATION OF PLATES 9-11

Abbreviations

- C.M., Constrictor muscle
C.T.N., Connective tissue nucleus
C.T.S., Connective tissue sheath
D., Duct
D.M., Dilator muscle
F.C., Funnel cell
G.C., Granular cell
G.L., Gland lumen
G.G.A., Granular gland anlage

G.S., Granular secretion
H.L., Horny layer
I.C., Intercalary cells
I.L.C., Inner layer of cells
M.C., Mucous cell
M.F., Muscle fiber
M.S., Mucous secretion
MX.C., Mixed cell
N.D.C., Non-differentiated cells
N.M.C., Nucleus of muscle cell
N.M.E., New mucous epithelium
O.L.C., Outer layer of cells
R.C., Replacement cells
R.G.A., Regenerating gland anlage
S.C., Schaltzellen
T.E., Transforming epithelium

All figures were drawn with the Abbe camera lucida.

PLATE 9

1. Median longitudinal section through a typical mature mucous gland showing duct, Schaltzellen, non-differentiated epithelial cells, mature mucous cells, discharging mucous cells, connective tissue sheath, muscles, epidermal replacement cells, intercellular bridges, horny layer. Ehrlich's haematoxylin and eosin. ($\times 660$).
2. Median longitudinal section through a typical granular gland; basal cell walls faintly visible, walls of upper cells completely disintegrated; secretion is being expelled. Horny layer is not shown. Ehrlich's haematoxylin and eosin. ($\times 660$).
3. Slightly tangential section, cut just below epidermis, to show arrangement of Schaltzellen, and muscle fibers. Mallory's connective tissue stain. ($\times 660$).
4. Section cut parallel to outer surface of epidermis, just above the corium, to show the funnel cell, replacement cells, dilator and constrictor muscles and nuclei. Mallory's connective tissue stain ($\times 1333$).
5. Longitudinal section through the epidermis, cut just to one side of the duct, to show epidermal muscle cells (large central pyramidal basal cell, just above corium), funnel cell and horny layer lifted from the duct. Mallory's connective tissue stain ($\times 1333$).

PLATE 10

6. Cross section through a mucous gland to show muscle fibers, mucous cells, transformation of mucous cells into granular cells; discharging mucous cells. Mallory's connective tissue stain ($\times 660$).
7. Similar gland to preceding one to show a more advanced stage of transformation of mucous cells into granular cells. Mallory's connective tissue stain ($\times 660$).
8. Functional mucous gland transforming into a granular gland; longitudinal

section. Cells of upper section of gland completely changed to a granular secretion; several cells on the right in the process of transformation—mucous cells. Mallory's connective tissue stain ($\times 660$).

9. Longitudinal section through a regenerating mucous gland to show complete disintegration of old mucous cells in the upper part of the gland, with new non-differentiated cells in this region. Lateral cell walls present in other gland cells; gland cavity completely filled with secretion. Mallory's connective tissue stain ($\times 660$).
10. A more advanced stage of a regenerating mucous gland; no indication of old mucous epithelium, entire gland cavity filled with mucous secretion. Upper section of gland shows new mucous epithelium and secretion differentiated. Mallory's connective tissue stain ($\times 660$).
11. Median longitudinal section through a gland to show ingrowth of granular epithelium to replace mucous epithelium—Mucous epithelium is being pushed from the gland wall. Mallory's connective tissue stain ($\times 660$).
12. The same gland cut two sections beyond the last. Mallory's connective tissue stain ($\times 660$).

PLATE 11

13. Section through a regenerating granular gland, cut just to one side and partly through the duct. Shows gland anlage in early stage of development, without a lumen. No differentiated cells in the upper part of anlage; transforming mucous cells, granular cells, and some mixed cells. Mallory's connective tissue stain ($\times 660$).
14. Longitudinal section through a regenerating granular gland; anlage differentiated into an outer and an inner layer of cells—becoming granular—cell walls are gone in the old gland; larger granules near the upper pole of old gland, smaller ones at the base; muscle and muscle nuclei at gland periphery. Ehrlich's haematoxylin and eosin ($\times 660$).
15. More advanced stage of a regenerating granular gland; outer and inner layer of cells in gland anlage, some cells are mucous, some are mixed, lumen in gland anlage. Cell walls of old gland have disintegrated, but an indication of their former presence is seen. Mallory's connective tissue stain ($\times 660$).
16. A more advanced stage in a regenerating granular gland. Gland anlage has differentiated into an almost fully developed gland with a large lumen; cells of the two layers of the anlage are of practically the same size; some are mucous, mixed, granular. No indication of cell walls in old gland—a syncytial condition of the old gland epithelium. Mallory's connective tissue stain ($\times 660$).
17. Cross section of a regenerating granular gland to show muscles of gland; gland anlage with narrow, non-differentiated inner layer of cells, lumen, tall cylindrical differentiated outer cells; granules of old gland are widely scattered in the gland cavity. Mallory's connective tissue stain ($\times 660$).

PLATE 9

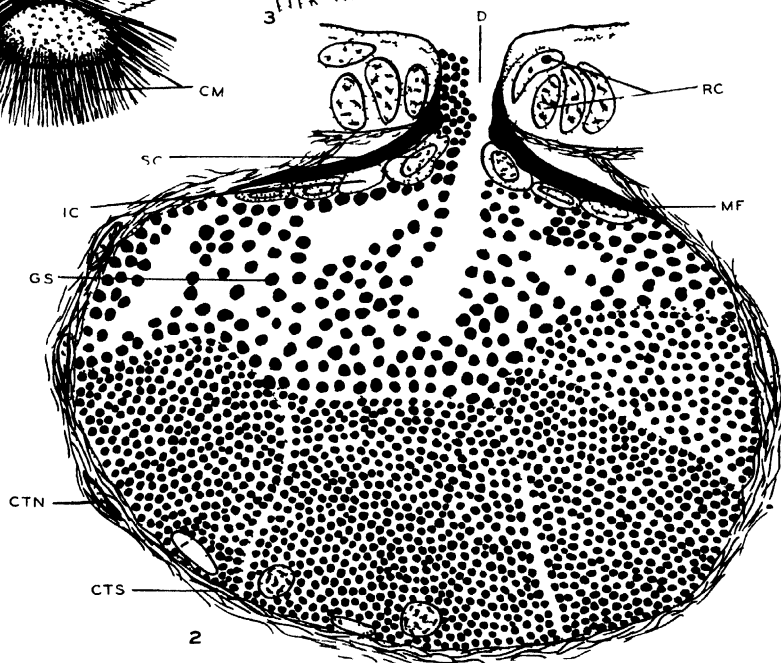
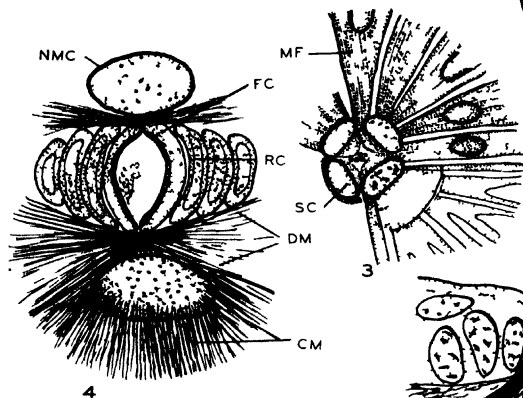
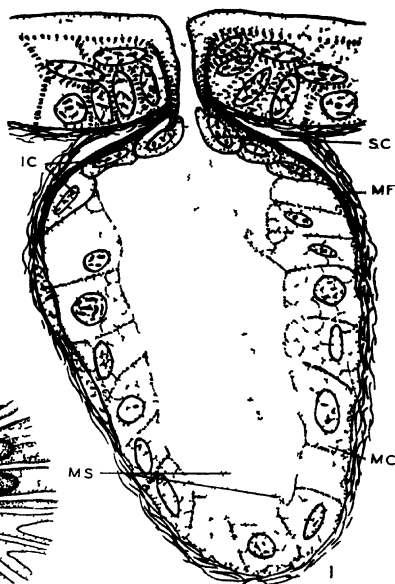
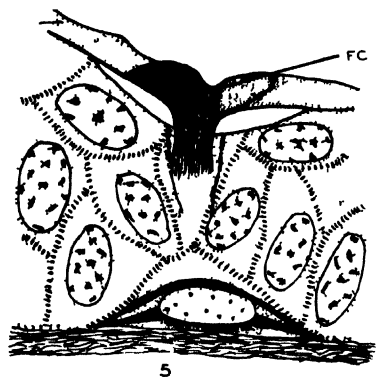


PLATE 10

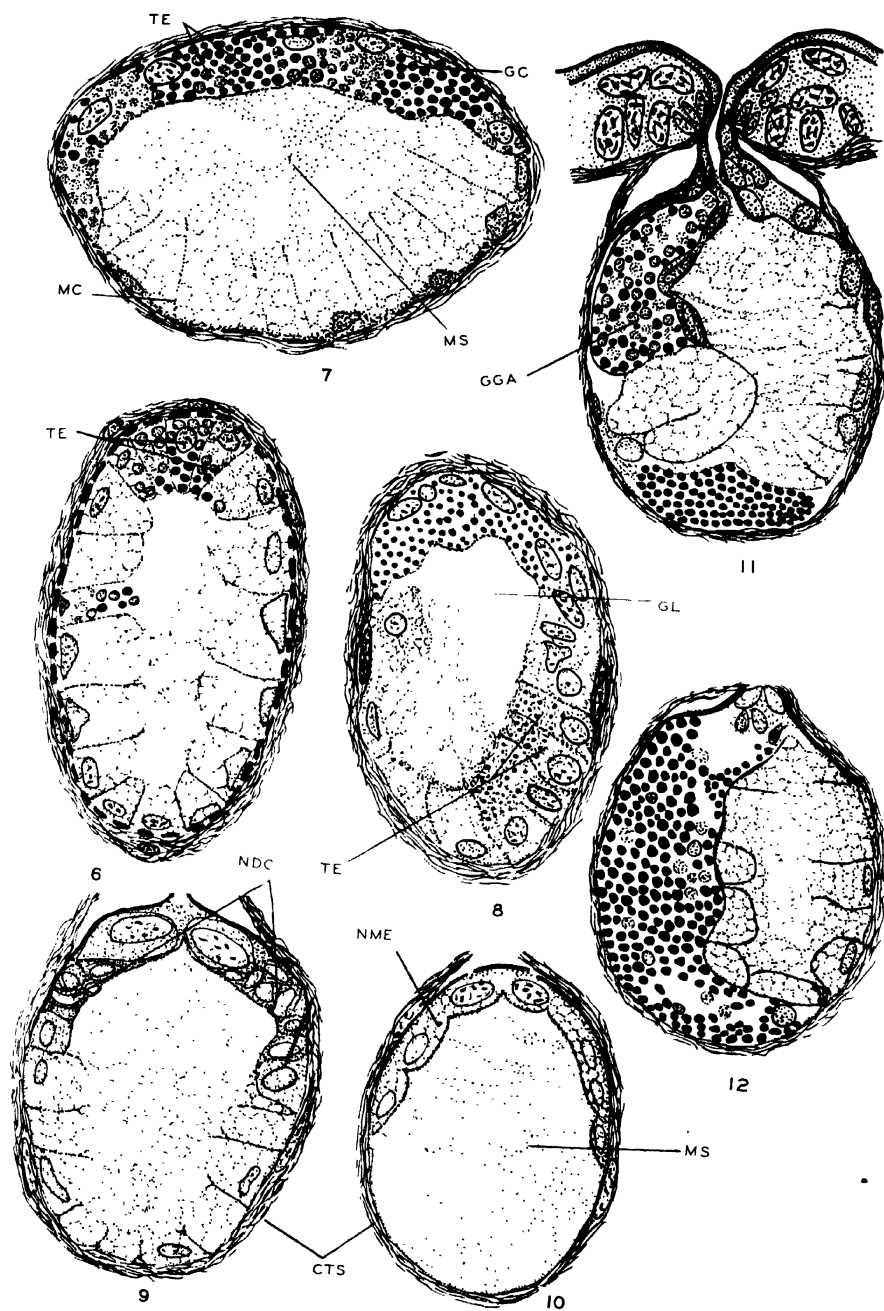
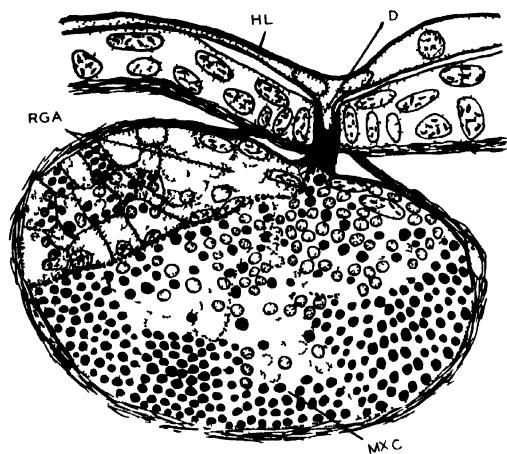
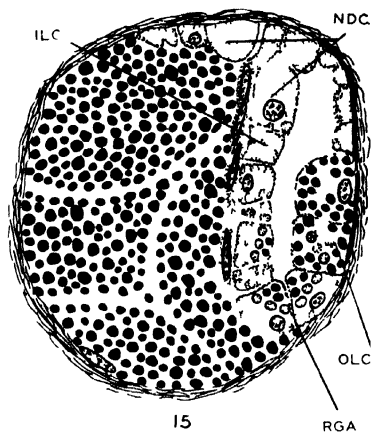


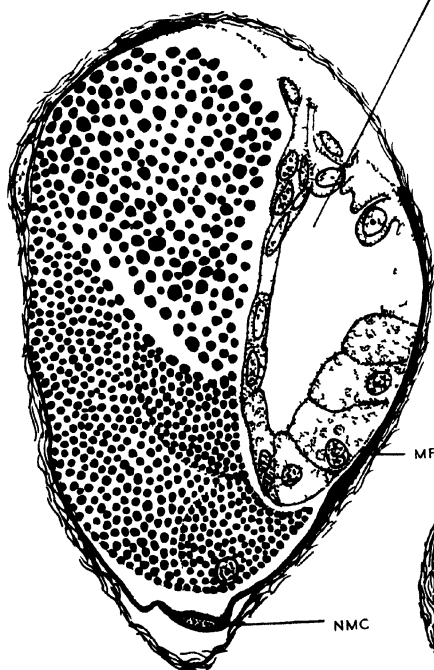
PLATE 11



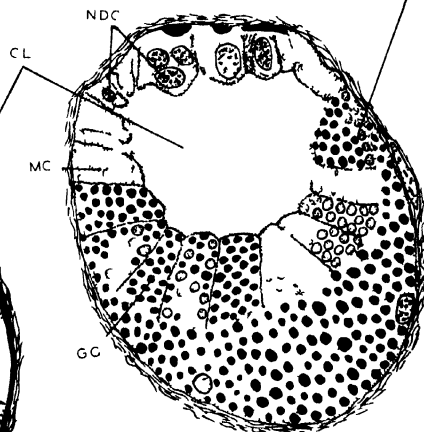
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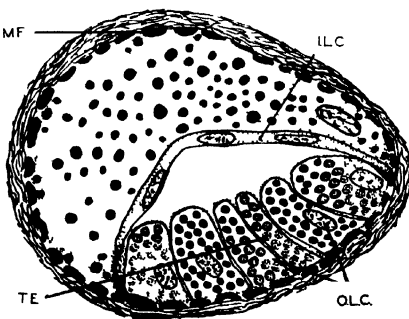
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17

TECHNIQUE IN THE PREPARATION OF CHICK EMBRYOS FOR CLASS USE

By C. D. BEERS

Special comment on the preparation of chick embryos for class use may seem on first thought superfluous, in view of the numerous methods already at the disposal of teacher and technician (e.g., the use of filter-paper rings, fixation on cover slips and various methods of pinning out on wax). But in my experience each of the methods described in the standard technique books offers in practice unforeseen difficulties which must be circumvented in each case by the ingenuity of the preparator. Of the various procedures that have been tried in recent years in this laboratory, the following seems to offer the minimum in technical difficulty and in loss of specimens through manipulative injury. While I recognize that there is little in the procedure that is fundamentally new, I have seen no mention of certain of the more useful details that follow, and a published record of the procedure in its entirety seems desirable as an aid to others.

Primitive-streak stage, incubation age 17 hours. This stage is undoubtedly the most difficult to prepare, not only because of its small size and extreme delicacy but because of the tendency of the vitelline membrane and the yolk to adhere tightly to the blastoderm. Embryos of this age may be fixed satisfactorily as follows: The egg is opened in the usual way, i.e., by picking away one side of the shell with forceps or by cutting around the shell in a plane parallel to its long axis, and the yolk mass is immersed in a dish of warm salt solution (0.9% NaCl, temp. 38°C.). The innermost layer of albumen usually adheres to the yolk at this stage; it must be picked away with forceps. With sharp scissors cut about three-quarters of the way around the blastoderm just at the periphery of the developing area *opaca vasculosa* not beyond, for beyond the periphery the vitelline membrane adheres almost inseparably to the developing blastoderm.

Some of the yolk invariably pushes through the 270°-cut just made and lifts up the free edge of the blastoderm. This almost always causes the vitelline membrane to curl slightly toward the attached edge of the blastoderm and to free itself from the latter to some extent. When

the vitelline membrane is visibly free from the blastoderm, complete the cut around the edge of the embryo and, using a teaspoon, transfer it to a dish of fresh salt solution.

As a rule a layer of yolk 2 to 4 mm. in thickness adheres closely to the under surface of the blastoderm. To remove it, turn the blastoderm so that its yolk surface is uppermost, and with a pipette gently wash away the yolk. This operation is time-consuming and requires the utmost in patience and attention. While the so-called nucleus of Pander beneath the area pellucida is easily removed, the remaining yolk adheres tenaciously to the area opaca. Therefore, the stream of salt solution must be directed always toward the periphery, for if it strikes the center forcibly, the blastoderm will surely tear. When nearly all of the yolk has been washed away, turn the blastoderm with its ectodermal surface uppermost, take hold of its edge with fine forceps and draw it onto a clean slide under salt solution. Using the slide as a lifter, remove the embryo from the solution. The embryo, now with its endodermal surface next to the slide, flattens out slightly, but not excessively, as it is removed. With a pipette take up the excess of salt solution from the slide, but do not attempt to wipe the slide dry with filter paper. Bring the end of a pipette filled with Bouin's picro-acetic-formol very near the center of the blastoderm and carefully flood the center with fixative in such a way that the fixative spreads from the center to the periphery. Under these conditions the blastoderm, particularly in its peripheral region where there is still some yolk, adheres closely to the slide. After a minute transfer the entire slide to a dish of Bouin's fluid and fix for 2 hours (longer does no harm).

If the excess of salt solution is removed with filter paper up to the edge of the blastoderm, the fixative, in flowing from the center to the periphery, also flows under the edge of the blastoderm and thus floats it off the slide.

After fixation, transfer the slide with attached embryo to 50% alcohol for 2 hours (Coplin jar), then to 80% alcohol (changed daily) for several days to wash out the picric acid, and finally to 95% alcohol for 2 days to complete the hardening and to keep the embryo from curling when it is later taken off the slide.

To remove from the slide, transfer to 80% alcohol for one hour, then to 50% for an hour, and with a safety-razor blade cut the embryo away from the slide under 50% alcohol. In this operation the edge of the blade must be kept pressed closely to the slide to avoid injury to the

endoderm. Transfer the detached embryo to 80% alcohol for an hour and preserve in 90% alcohol. The removal of the embryo in 50% alcohol rather than in 95% reduces the possibility of injury, since embryos stick less tightly to the slide and are less brittle in 50% alcohol.

More than five dozen embryos have been prepared by the foregoing method and sectioned in the last three years in this laboratory with no appreciable injury to the endoderm.

Incubation ages 21 hours to 4 days. Embryos of these ages are pinned out with No. 0 or No. 1 stainless steel insect pins on black, weighted paraffin blocks which are prepared as follows: Have ready some pieces of sheet lead about 3 cm. square and 2 mm. thick. Stir about 5 cc. of lamp-black into about 200 cc. of melted paraffin wax (m.p., 56°C.). Smear the inside of a clean Syracuse watch glass with a film of pure glycerine. Fill the watch glass to a depth of about 3 mm. with the paraffin mixture, put the piece of lead into the watch glass, fill completely with paraffin and, as soon as the surface hardens slightly, cool in running water as in watch-glass imbedding. Upon cooling, the block contracts away from the watch glass and is easily removed. Such blocks present a slightly concave upper surface and are heavy enough to sink in water.

Open the egg in the usual way and transfer the yolk mass to a dish of salt solution. Cut around the blastoderm 2 to 5 mm. beyond the area vasculosa, depending on the age of the embryo, and free the blastoderm from the yolk. The vitelline membrane and the yolk are readily detached from the embryo after the 20th hour. Fill a second dish with warm salt solution and immerse a paraffin block in it. With a spoon transfer the embryo to the second dish and pin it out under salt solution on the block. In pinning, allow for some shrinkage during fixation and washing in alcohol. A Camel's hair brush is useful in unrolling the edges of the blastoderm during pinning. Carefully remove the block from the salt solution, take up the excess with a pipette, and flood the embryo with Bouin's fluid. Then immerse the block completely in the fixative for 4 hours or longer.

After fixation transfer the block to 50% alcohol for 2 hours and then to 80%, changed daily, for several days. Finally, harden in 95% for 2 days, remove the embryo from the block and preserve it in 90% alcohol. Unless the embryo is hardened in 95% alcohol before removal from the block, it will curl later upon preservation in 90% alcohol.

Primitive streaks may also be fixed by this method, but their small size increases the technical difficulties, in that even the fine points of No. 0 insect pins may injure the embryos by distorting the cell layers.

After a thorough washing in running water, the blocks and pins are ready for use again. The very fine points of stainless steel insect pins and their capacity to resist chemical action make them ideal for pinning out chick embryos, though some types of pins have brass heads which should be paraffined before use.

A convenient lifter for transferring the blocks from one solution to another may be made from a teaspoon. Flatten the bowl of the spoon by hammering and bend it so that it forms an angle of about 120° with the handle. Coat the spoon with paraffin to protect it from the fixing and washing fluids.

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OBSERVATIONS ON THE DEVELOPMENT AND CYTOLOGY
OF THE SEXUAL ORGANS OF *THRAUSTOTHECA*
CLAVATA (DE BARY) HUMPH.

By LELAND SHANOR

PLATES 12 AND 13

INTRODUCTION

Thraustotheca clavata (de Bary) Humphrey, though widely distributed, is one of the rarer members of the Saprolegniaceae. It was first discovered by de Bary in a collection of algal material from Vendenheim, near Strassburg, Germany, in December, 1880, and named by him *Dictyuchus clavatus*. Büsgen (4) in 1882, incidentally published the first description of this fungus as *Dictyuchus clavatus* de Bary sp. nov. when he was making a study of sporangium formation in various species of the Phycomycetes. De Bary (2) refers to it again as *Dictyuchus clavatus* in his text book where he mentions this fungus as an example of endogenous spore formation. In a paper by de Bary (3) which was published in 1888, after his death, and edited by Solms-Laubach, the latter suggested the possibility of this fungus not belonging in the genus *Dictyuchus*. Fischer (19) in 1892, in his work on the Saprolegniaceae, also felt that the strikingly different characteristics of this fungus should place it in a genus separate from *Dictyuchus*, but he did not remove it from this genus. In 1892, Humphrey (27) thought it sufficiently different from other members of the genus *Dictyuchus* to be separated, so he placed it in his new monotypic genus, *Thraustotheca*, even though he had never had the fungus to study. Von Minden (30) reports it again from Germany in 1912.

In America, collections of *Thraustotheca clavata* have been recorded several times, most often by Dr. Coker and his students. It was first reported in this country by Coker and Hyman (10) from Chapel Hill, North Carolina, in 1912. Weston (42) found it near Great Barrington, Massachusetts, in 1913, and reported it in his paper which appeared in 1918. Harvey (24, 25) reports collections from the soil in Chapel Hill in 1925, and from soil collections from Wisconsin, New York, and Kentucky in 1930. Coker and Braxton (9) report finding it in

Chapel Hill, March 1926, and in Haywood County, North Carolina, in July and August of the same year. Couch (14) reports collections of *Thraustotheca clavata* from Cold Springs Harbor, L. I., New York, and from Abbey Green, Jamaica, B. W. I., in 1926.

The material from which this study was made was obtained by the author in a water collection from a spring in the College Woods at Maryville, Tennessee, November 27, 1935. Shortly after this collection was made, *Thraustotheca clavata* was isolated from other material in this collection and identified in the Botanical Laboratory of the University of North Carolina.

The question of fertilization in the Saprolegniaceae had been much disputed for a long time. Among the early investigators, Pringsheim (33, 34) argued for the necessity of fertilization in this group and felt that it took place at least in some species, even though he did not actually observe it to occur. Cornu (13) believed that fertilization occurred in *Achlya polyandra* Hildb. and in *Achlya racemosa* Hildb. and thought that fertilization in other species was extremely probable. De Bary (2) at first thought that it might occur, but later concluded that the antheridia, although present in some members of the family, were now functionless. Humphrey (27) and Hartog (21, 22, 23), both working with stained material, believed as de Bary did about the matter, but described a pairing and fusing of the nuclei in the oogonia of some species. Davis (17, 18) likewise believed that fertilization was not found in this family.

Trow (37, 38, 40, 41) was the first of the early cytological investigators of this group to believe and to demonstrate that fertilization took place in the Saprolegniaceae. He described nuclear division in the antheridia and oogonia and claimed that the supernumerary nuclei degenerate rather than fuse in pairs as had been described by Hartog and Humphrey. His observations of degenerating nuclei in the oogonia here were strongly questioned. In order that he might be more certain that he had observed degenerating nuclei, he made a study (39) of *Pythium ultimum*, a member of a family where the degeneration of nuclei was known to occur. He then made further studies on two species of *Achlya*, and as a result of these observations, showed clearly that nuclei degenerate in the oogonia. He asserted then that he was more certain than ever that fertilization occurred, not only in *Saprolegnia dioica* de Bary, *Saprolegnia mixta* de Bary, and *Achlya americana* Humph., as he had believed before, but also in *Achlya polyandra* Hildb. and *Achlya de Baryana* Humph. He held to this view in spite of the

severe criticism and opposition of several of the investigators of that time, especially of Hartog. Trow (40) also claimed that there was evidence of a second mitosis in the oogonium, which he thought was probably reductive. This second mitosis has not been observed since and later investigators believe that the reduction takes place in the germination of the egg.

Since this early controversy, however, fertilization has been definitely demonstrated in several species in this family. It has been shown to occur in *Saprolegnia monoica* (Prings.) de Bary, by Claussen (6); in *Aphanomyces laevis* de Bary, by Kasanowsky (28); in *Achlya polyandra* de Bary, by Mücke (31); in *Achlya racemosa* Hildb., by Carlson (5); in *Achlya colorata* Prings., by Patterson (32); in *Achlya hypogyna* Coker and in *Brevilegnia diclina* Harvey, by Cooper (11, 12); in *Leptolegnia caudata* de Bary, by Couch (15); and in *Saprolegnia mixta* de Bary, by Mäkel (29). The evidence also indicates that fertilization takes place in oogonia of *Saprolegnia ferax* (Gruith.) Thuret that are contacted by antheridia (Höhnk, 26), and although not actually observed, Raper (35) presents considerable evidence of its occurrence in *Achlya bisexualis* Coker.

In *Thraustotheca clavata* it has been thought that fertilization takes place, but the cytology of this form has not been worked out. Coker (7) in his monograph of the Saprolegniaceae, in summarizing the work done on this plant, says that fertilization has never been observed, but that the antheridia become empty during the ripening of the eggs. Weston (42) says regarding his observations: "It is worthy of note that the fertilizing tubes were never seen to penetrate the oospheres, nor was any sudden passage of material down the tubes observed." He also states that, without exception, the oogonial contents never separate into oospheres until the attachment of the antheridia to the oogonium had taken place.

The purpose of this study is: (1) to follow the development of the sexual organs in living material, and (2) to make a cytological study of the development of the antheridia and oogonia in fixed material.

MATERIALS AND METHODS

Pure cultures were obtained from the original collection by planting a single sporangium that had been thoroughly washed in sterilized distilled water on a plate of maltose-peptone agar. The cultures were then transferred from the agar to hemp seed in water. This was done by cutting out small pieces of the agar bearing the fungus and placing

on each of them in a Petri dish a half of a sterilized hemp seed. Hemp seed in water proved the most satisfactory media for carrying on stock cultures of *Thraustotheca*. The water used in these cultures was distilled water to which had been added a small quantity of animal charcoal. It was then filtered and sterilized in the autoclave at twenty pounds pressure before being used. The stock cultures were kept growing in Petri dishes in water on hemp seed from November 1935, until January 1937, when this study was begun. Fresh hemp seed were added whenever necessary.

When the present study was begun, it was necessary to have cultures as free from bacteria as possible. The method described by Raper (35) was found to give excellent results, if the blocks of agar were taken out shortly after the hyphae had appeared outside the glass ring. If the transfer was delayed until much later, the cultures on agar were generally found to be contaminated with bacteria.

On hemp seed, young cultures of *Thraustotheca clavata* produced a very vigorous growth with numerous sporangia. Later oogonia and antheridia were produced, but these were quite frequently so hidden by the hyphae and sporangia that their development was difficult to observe. To get cultures that produced numerous oogonia and antheridia without such a vigorous growth of hyphae and sporangia, flies and termites that had been preserved in eighty percent alcohol were used. Of these two, the termites were found to be the more satisfactory because they produced a less vigorous growth and at the same time, numerous oogonia and antheridia.

For cytological studies, cultures growing on termites were killed and fixed when they had reached the desired stage in development, as was determined by a microscopic examination. Each culture, together with the termite, was lifted carefully and dropped into the fixing solution. As killing and fixing agents hot corrosive sublimate, formalin-acetic-alcohol, a chromo-acetic acid solution, Fleming's weak solution, Raper's (35) chromic acid-formalin solution, Merkel's fluid, Carnoy's fluid, and the Chicago formula as described by Chamberlain, were tried. Fixation was usually for from twenty-four to thirty-six hours. Raper's chromic acid-formalin solution was found to give better results than the others when followed by the Gram's stain as described by Couch (15), and was used almost entirely throughout this study.

After fixation, material was washed for twenty-four hours in running tap water, dehydrated in the usual manner, and imbedded in 53-56 degree paraffin. Sections were cut 5, 6, and $7\frac{1}{2}$ μ thick with a Spencer

rotary microtome. Sections were then stained on the slide with either Heidenhain's iron-alum haematoxylin or Gram's stain. Gram's counter-stained with orange G was also tried, but was not so satisfactory as the Gram's without the counter stain. The hematoxylin was not satisfactory because small granules in the cytoplasm held the stain so greedily that they rendered the details of nuclear structures indistinct. Material fixed in solutions containing osmic acid was also unsatisfactory because certain granules again would interfere in the same way. Trow (40) had the same difficulty with these granules, when using a haematoxylin stain, when working on *Achlya de Baryana* and referred to them as microsomata.

In the latter part of this study when particular attention was attracted to the astral rays, the time schedule from the iodine to mounting in balsam as given by Couch (15) was found to be too long because the alcohols and clove oil would remove too much of the gentian violet from the cytoplasm. Therefore, after the iodine treatment and the wash which follows it, as much of the water as could be removed with filter paper was drawn off and the slides then passed as quickly as possible through two changes of absolute alcohol, clove oil, cedar oil, and then into xylol. The clove oil and cedar oil were sometimes omitted, this treatment as a rule giving results equally as good as the other.

The gentian violet used in the Gram's technique was Gentian Violet (Crystal Violet) manufactured by The National Aniline and Chemical Co., Inc., New York.

DEVELOPMENT OF THE SEXUAL ORGANS IN LIVING MATERIAL

The development of the sexual organs of *Thraustotheca clavata* in living material has been described and figured in considerable detail by Coker and Hyman (10) and by Weston (42). My observations agree essentially with those of these observers and no additional figures seem necessary.

Young oogonial branches were found numerous in cultures grown on termites that were three or four days old. Antheridial branches were also numerous and these twined about among the hyphae and were often noticed to branch.

On my termite cultures, a number of oogonia were found that had formed oospheres without the attachment of antheridia. Both lateral and intercalary oogonia whose eggs were developing parthenogenetically were observed quite commonly toward the periphery of these cultures,

but no mature eggs were ever found that had developed in this manner. Weston (42) was of the opinion that oospheres were never formed until after the attachment of antheridia had taken place. In oogonia in his cultures that were not contacted by antheridia, oospheres were not formed even when he followed the method successfully used by Trow (38). Coker and Hyman (10) say nothing of this peculiarity, but their figure 8 shows an oogonium in which the oospheres have formed without the attachment of antheridia to the oogonium.

The development of the sexual organs was followed through at several different times. Cultures grown on termites were used for most of these observations because of the greater ease with which the developing oogonia could be followed without the interference of a dense growth of hyphae. The period of development from the formation of the oogonial initials until the eggs were mature took about four days.

Young stages in the oogonial development are found most numerous from early morning until about noon. Very early the lateral branches which will bear oogonia can be distinguished because they are becoming more densely filled with protoplasm. Soon the tip begins to swell and to become rounded to form the young oogonium. The protoplasm in the oogonium at this stage is densely packed and appears to be more or less homogenous; the small oil globules are hardly distinguishable.

Soon a central vacuole makes its appearance and a cross wall is laid down which separates the oogonium from its stalk. Usually by this time one or more antheridia have become attached to it. The oil globules in the oogonium can be faintly seen by the time the vacuole is fully formed. The vacuole soon begins to send out projections which pierce the protoplasm, separating it into several cone-shaped masses, the number depending upon how many eggs are to be formed. When this cleavage has been completed, these cone-shaped masses of protoplasm round up to form the oospheres. These young oospheres contain many small oil globules which are now readily observed. Coker and Hyman (10) found these globules arranged more to one side of the oosphere at this stage in development, but in my observations they seemed to be scattered indiscriminately all through the oosphere.

Each oosphere now has a thin membrane formed around it, and it is at this time that fertilization takes place. Fertilization is described under the discussion of the antheridium. After fertilization has taken place, the oil globules now begin to fuse to form a single eccentric, large globule. The wall of the egg becomes much thicker by a deposit of cellulose. In this condition the eggs are mature.

Because of their twining and branching habit, many antheridial branches are able to reach oogonia. Soon after the attachment of a branch to an oogonium takes place, protoplasm containing many oil globules flows into the tip of the branch. After a short time many of these oil globules seem to flow back into the basal part of the branch or into the hyphae from which it arose, leaving only a few in the tip of the branch. A cross wall soon forms across the tip of a branch that has become attached, thus forming an antheridium. The oil globules in the antheridium tend to fuse to a certain extent and several denser greyish appearing places are evident at this time. These are thought to be nuclei because of their size and greyish appearance. No further change was observed in an antheridium from this time until after the oospheres had rounded up in an oogonium.

After the oospheres have been formed in an oogonium, short fertilization tubes are sent into the oogonium by the antheridia. These often grow past the edge of the egg and are then lost from observation. Protoplasm from the antheridium flows into these tubes. Fertilization takes place very soon after the eggs have assumed their definite form. The actual passage of material from the tubes into an oosphere was not observed. One oosphere was observed about the time fertilization would take place in which what appeared to be the path of a male nucleus from the end of a tube to near the center of an oosphere was seen. A search for other such stages proved unsuccessful.

A slow disintegration of the protoplasm which remains in an antheridium or the fertilization tubes seems to take place, for particles of protoplasmic matter and oil globules could be observed for some time. There did not appear to be any emptying of the protoplasm from the antheridium into the oospheres during fertilization. This disintegration process progresses until by the time the eggs are ripe, the antheridia appear to be entirely empty.

OBSERVATIONS ON FIXED MATERIAL

Resting vegetative nuclei in the hyphae are found to vary considerably in shape. This same observation has been reported by Smith (36) in *Saprolegnia dioica* and described there in detail in relation to direct division of nuclei in vegetative hyphae.

Spindle-shaped nuclei are found in the flowing protoplasm in the hyphae. In positions where the streaming would not take place so rapidly, the nuclei become more spherical in shape. Spherical nuclei are found in young sporangia and in early stages in the development

of the sexual organs. As the nuclei are carried into the young oogonia by the streaming of the protoplasm, they show the characteristic spindle-shaped appearance, but upon entering the young oogonium proper they become spherical in shape (fig. 1). Smith (36) ascribes the spindle shape of nuclei in flowing protoplasm here to a tension or strain within the semi-liquid cytoplasm. It seems likely that this interpretation is correct. In *Thraustotheca clavata* nuclei assuming this elongated form are not found except in parts of the hyphae where the protoplasm was most likely streaming at the time when the material was killed and fixed.

A large number of nuclei are carried into young oogonia by the streaming of the protoplasm. Both young antheridia and oogonia are multinucleate, but the number of nuclei in antheridia is relatively small; probably seldom more than eight or ten. As the nuclei arrive in the tip of an oogonial branch they become spherical as has already been stated. The number of nuclei which are thus carried into a young oogonium before the cross wall is laid down varies considerably with the size of the oogonium. The number probably ranges somewhere between thirty-five and nearly one hundred.

The vacuole begins to appear in young oogonia before the cross wall separates an oogonium from its stalk. The vacuole gradually becomes more spherical, enlarges somewhat, and takes up a central position in the oogonium. Antheridia have usually become attached to oogonia by the time this stage is reached.

Nuclei at this stage of oogonial and antheridial development are very much the same in size and in structure. Each nucleus at this time shows a distinct, large, deeply staining nucleolus (fig. 1a). About this nucleolus there is a nuclear plasm which is limited by a nuclear membrane. From the nucleolus to the membrane there radiate a few linin threads which are attached to the membrane and make up the nuclear network. Several darkly staining granules of chromatin material can be seen on the nuclear membrane and on the linin threads. Chromatin material on the nuclear membrane has been observed by several others who have worked on the cytology of the water molds. Besides the chromatin on the nuclear membrane, there can usually be identified at this stage a larger, darkly staining body, the centrosome. These structures show well in material stained with the Gram's technique. Claussen (6) and Mücke (31) have noted a centrosome on the membrane of resting nuclei at this stage. Höhnk (26) denies the pres-

ence of a definite centrosome at any time in the development of *Saprolegnia ferax*.

Regarding the structure here referred to as a nucleolus there seems to be some difference in the interpretation of its function. Hartog (21) regarded it as a chromatin body. Trow (37, 40) thought that it was a combination of a nucleolus and a chromosome. Davis (17), Claussen (6), Mücke (31), and Carlson (5) regarded it as a nucleolus. Patterson (32) agreed with Trow on the latter's interpretation of this body.

At about the stage in the development when the cross wall which cuts off the oogonium from the stalk is formed, a number of these oogonial nuclei were found in various stages of degeneration. Degeneration of these nuclei seemed rather rapid, for by the time the spindle is formed in the mitosis of those nuclei which remain, they can be detected only as deeply staining granules scattered in the cytoplasm (fig. 4). A short time later they cannot be detected at all (fig. 9).

The nuclei which remain in the oogonium enlarge considerably before the mitotic division takes place. In early prophase, the chromatin material on the nuclear network and on the nuclear membrane becomes quite thickened (fig. 2). The centrosome is often quite conspicuous at this time. In later prophase stages, the threads themselves also seem to become thickened to give the threads a more or less uniform appearance. Carlson's (5) figure 13 of *Achlya racemosa* shows essentially the same structure as nuclei of *Thraustotheca clavata* in this same stage. Later in the prophase the chromatin material seems to draw away from the nuclear membrane and to become clustered in the form of curious rod-like bodies in the equatorial region of the nucleus (fig. 3). The nucleolus loses its identity during the prophase and is thought to contribute to the chromatin of the spireme. This also seemed to be the fate of the nucleolus in nuclei of *Leptolegnia caudata* as observed by Couch (15). Carlson (5) believed that the nucleolus in nuclei of *Achlya racemosa* decreased in size and finally disappeared all together. She does not explain how this takes place or explain its fate other than to say that it disappears. I have been unable to find stages showing the formation of the spindle and the passage of the nuclei from the prophase to the metaphase.

Nuclei in the metaphase show a definite clearly defined spindle with the chromatin aggregated in the equatorial region (figs. 4 & 6). The long axis of the spindle is almost always nearly parallel to the original wall in exact median sections of oogonia. In well stained sections there appear to be three distinct fibers. A fourth one shows faintly in some

preparations and it seems that this one was probably just obscured by one of the others where three fibers were seen. There is a distinct centrosome at each pole of the spindle, from which radiate astral rays (figs. 4 & 6). The astral rays do not stain here as clearly as they do in later stages, but nevertheless, they are readily discernible. Astral rays associated with metaphase nuclei have been observed in both antheridia and oogonia (figs. 4-6). Claussen (6) has observed them in the antheridia of *Saprolegnia monoica* and it is a well known fact that they occur in the oogonia of several species of the water molds. The origin of the two centrosomes found here from the one found in earlier stages has not been determined.

Some of the nuclei at the metaphase show the spindle to be situated within what appears to be a nuclear membrane; others show no such structure present. Because some of the nuclei begin to degenerate at this stage rather than later after the division is complete, it is thought that those in which no structure resembling a membrane appears are in early stages of degeneration. Mitosis in the antheridium does not often occur simultaneously with that in the oogonium, but rather takes place after mitosis in the oogonium has progressed somewhat or is almost completed (figs. 4 & 9). This has been the rule in all sections showing mitosis in the antheridia which I have observed. There seems to be only one mitosis in either the oogonia or antheridia of *Thraustotheca clavata*.

The chromosomes stain quite darkly with the gentian violet. They are very small and are variable in shape. Some appear to be slightly rod-shaped, others more spherical, and still others somewhat irregular in appearance. They tend to cluster together so closely that their number could not be determined with certainty. There are definitely more than three as described for *Achlya colorata* by Patterson (32) or from four to six as noted in *Achlya racemosa* by Carlson (5). Claussen (6) found from ten to fourteen in *Saprolegnia monoica* and Mäkel (29) reports from his studies on several species that the chromosome count was constantly eleven. In *Thraustotheca clavata* the chromosome number seems to correspond more nearly with the number observed by Claussen and Mäkel. The extremely small size of the nuclei and of the chromosomes in any of the water molds makes the problem of determining their number with any degree of certainty an extremely difficult one.

As the chromosomes split and start migration toward the poles, the membrane-like structure disappears (fig. 7). The chromosomes are drawn toward the centrosomes (fig. 8), but the daughter nuclei in the

oogonium do not form resting nuclei at this time. The chromatin material remains in a darkly staining mass a very short distance from the centrosome and is connected with it by a faintly staining beak process (figs. 9, 10, 11). Degeneration of all of the daughter nuclei except those which are to remain to function as female gamete nuclei in the oogonium now takes place (fig. 9). Degenerating nuclei in various stages of disintegration can be detected for a short time. These degenerating nuclei appear very similar to those described by Claussen (6) at this stage in *Saprolegnia monoica*. Degeneration of nuclei here is again very rapid, for even late stages were not observed in the egg origins.

Shortly after this mitotic division, the cleavage furrows from the central vacuole begin to push outward. As this occurs, female gamete nuclei whose centrosome and astral rays are not directed outward from the center of the oogonium seem to rotate so that their centrosomes and rays are directed toward the oogonial wall (figs. 9, 10, 11). This outward direction of the centrosome and astral rays at this time seems to be universal in this family for the figures of other observers bear this out. In my observations there was no departure from this rule. Couch (16) has also observed this to be the case in *Achlya orion*, *Achlya apiculata*, *Achlya caroliniana*, and in *Saprolegnia delica*. Claussen's figure 18 shows a very faint centrosome with the rays directed inward, but his other figures would lead one to believe that this is not typical. The astral rays at this stage become much longer and branched so that they are in intimate contact with the cytoplasm within a considerable radius of the centrosome and the oosphere nucleus. They can be traced even out into the bridges of cytoplasm which extend between the oosphere origins (fig. 12). The number of egg origins found in an oogonium varies. Oogonia containing a single large uninucleate egg have been observed as well as oogonia containing as many as ten eggs. The average number seems to range from four to six.

As the cleavage furrows push outward toward the oogonial wall, the egg origins become more and more cone-shaped (fig. 11). The astral rays at this stage, and especially during the rounding up of the oospheres, can be traced to the very margin of the oosphere cytoplasm (figs. 11, 13, 14). Some of these rays were seen to branch just as they were about to reach the margin. Claussen (6) was able to trace these rays to the margins of the oospheres in *Saprolegnia monoica*, but his figures do not show this clearly. At any rate, they must not have been nearly as distinct as those found in *Thraustotheca clavata*. After the oospheres have rounded up, a thin membrane is formed around each of

them and the astral rays become much shorter but remain distinct (fig. 15).

At about the time that the membrane is formed around the oospheres, fertilization tubes are sent into the oogonium from the antheridia (fig. 13). These tubes sometimes grow directly toward the nearest oosphere and at other times are observed to become quite long and to wind about in the oogonium before making contact with an oosphere. The former condition was the more frequently encountered. Protoplasm from the antheridium flows into these tubes as they elongate. The antheridial nuclei stain rather darkly at this stage and the chromatin seems to be massed together. The astral rays that were observed during the mitosis are not seen at this time.

As the fertilization tube reaches an oosphere it presses against the membrane and the end of the tube enlarges somewhat (fig. 16a). The end of the tube finally ruptures the oosphere membrane and a single male nucleus is released into the oosphere (fig. 16b). Other male nuclei in the tube and those in the antheridia which are not to function now degenerate. It has been noticed that an oosphere at the time of fertilization shows a vacuole-like region near the point of entrance of the male nucleus. This might be referred to as a fertilization spot. A somewhat similar condition has been shown by Couch (15) at the time of entrance of the male nucleus in *Leptolegnia caudata*. Carlson (5) also shows what appears to be a fertilization spot in *Achlya racemosa*. The fertilization tube has been observed to collapse somewhat after the male nucleus has been released into the oosphere (fig. 16b).

The male nucleus proceeds toward the female nucleus which is usually located near the center of the oosphere. As it passes along it leaves a path through the cytoplasm (fig. 17). Cooper (12) described and figured such a "penetration path" of the male nucleus in *Brevilegnia declina* and Couch (15) shows a very distinct one to be present in *Leptolegnia caudata*. The male nucleus finally takes up a position not far from the female nucleus near the center of the egg and there the centrosome with the astral rays again are seen associated with it. The female nucleus also shows astral rays to be still present (fig. 17). Soon after this the male and female nuclei become transformed into nuclei which resemble in structure resting nuclei (fig. 18). These nuclei now approach each other and lie in contact for some time (fig. 19). The membrane between the two finally disappears and fusion between the nuclei takes place (fig. 20). The zygote nucleus is slightly larger than either of the gamete nuclei and stains more darkly. It usually lies

near the center of the egg at this time (fig. 21). I have been unable to stain satisfactorily mature or germinating eggs, because structures in the position of the oil globules in living material are, at these stages, so large and stain so darkly that other structures are obscured.

AN OBSERVATION ON YOUNG ZOOSPORES

A short time before the study on the development of the sexual organs was completed, a study of sections of the zoosporangia which had been sectioned was made. Astral rays associated with a centrosome in zoospores was observed in sections of material fixed in the chromic acid-formalin solution and stained by the Gram's technique. A large number of sporangia were then examined which revealed the presence of this centrosome and astral rays in the zoospores. Unfortunately, when material is fixed for the study of the development of the sexual organs, only late stages in zoospore formation in the sporangia are obtained. The reason for this is because the antheridia and oogonia are not usually produced in abundance until after most of the stages in the early formation of the zoospores are passed. The complete cytology of spore formation in *Thraustotheca clavata* has not been worked out, but a single figure (fig. 22) of a sporangium after the spores have been delimited is here included to show the structure of the spore nuclei and the appearance of the centrosome and astral rays.

DISCUSSION

The cleavage of the oospheres in the Saprolegniaceae is generally attributed to the cleavage furrows from a central vacuole. Claussen (6) regarded them in this respect as the active agents in oosphere delimitation. Davis (17) believed that at least a part of the activity of the "balling" of the egg origins was due to a "coenocentrum." This Davis described as the "morphological expression of dynamic activities in the oogonium, and especially in the egg origins at the time when they were differentiated." Claussen (6) disagrees with Davis on this matter for he feels that Davis is wrong in confusing such a structure as is found here with the coenocentrum which is found in the Peronosporaceae.

It is a striking fact that in all egg origins and newly formed oospheres of *Thraustotheca clavata* the centrosome constantly occupies a central position in each protoplasmic mass. From this central body there radiate ramifying astral rays which can be traced to almost the margins of the egg origins. Because these rays are so long and come so

very close to, if not reaching the margins of the oospheres, it is felt that the central body, or centrosome, and the astral rays contribute to the cleavage of the eggs.

This possibility is further brought out when one considers the function of the astral rays in the formation of the ascospores in the Ascomycetes. Harper (20) and Bagchee (1), among others, have given us detailed accounts of the function of the astral rays in spore delimitation in this group. In the Ascomycetes the centrosome, at this time in particular, is definitely the center of the activity of the nucleus as interpreted by Harper. He finds this central body associated with the nucleus at all times. The astral rays found in the Ascomycetes not only serve as the agents for the delimitation of the ascospores, but also later these structures give rise to the plasma membrane of the spores. Such a relationship between the astral rays and the oosphere membrane has not been observed in *Thraustotheca clavata*.

When one considers these facts, it would not seem at all impossible that the astral rays found associated with the female gamete nuclei in the oogonium may function in oosphere formation. It is obvious that they do not "cut out" spores as is the case in the Ascomycetes, but their function in the cleavage and rounding up of the egg origins in this plant seems evident in the light of these observations.

Since the size of the eggs in *Thraustotheca clavata*, except in cases where an oogonium contains a single large egg, is relatively constant, some internal force in the region of the egg origins must function in the apportionment of the oogonial cytoplasm that goes into the formation of each of these, besides the part taken by the cleavage furrows. The length of the rays and the position of the centrosome before the cytoplasmic bridges between egg origins are broken also would seem to add weight to this conclusion. The importance of the cleavage furrows in the delimitation of the oospheres is not denied. However, it is felt that these are not the only agents that function in the delimitation and rounding up of the oospheres. The centrosome and the astral rays also seem to function in this process in *Thraustotheca clavata*.

SUMMARY

1. The observations on living material conforms in general with those of other observers.

2. A combination of a chromic acid-formalin killing and fixing agent with Gram's staining technique is found to give very good results for demonstrating such cytoplasmic structures as the acromatic figures.

3. The general cytology of *Thraustotheca clavata* is essentially in accord with the observations of others who have worked on the Saprolegniaceae.

4. Fertilization is shown to take place in *Thraustotheca clavata*, thus adding another genus and another species in the Saprolegniaceae in which fertilization has been demonstrated.

5. In the cleavage of the egg origins, the centrosome and the astral rays are found to be directed outward in all cases. The centrosome always occupies a nearly central position in the cytoplasmic mass of the egg origins and of the young oospheres. The astral rays are shown to extend out to the margins of the young oospheres and out into the cytoplasmic bridges between egg origins.

6. Astral rays associated with a central body are shown to be present in young zoospores. The cytology of spore formation has not been worked out.

7. In view of the position of the centrosome and the length of the astral rays during the formation of the oospheres in *Thraustotheca*, a relationship between the function of the acromatic figure here and of that in the Ascomycetes is advanced.

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The writer wishes to express his sincere gratitude to Dr. W. C. Coker and to Dr. J. N. Couch for their encouragement and helpful suggestions during this study. I also wish to acknowledge my indebtedness to Dr. J. E. Adams for suggestions on staining technique and to Mr. Don Ritchie and Miss Laurie Stewart for assistance and suggestions at various times.

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LITERATURE CITED

1. BAGGEE, K. 1925 Cytology of the Ascomycetes. *Pustularia bolarioides* Ramsb. Ann. Bot. 39: 217-226, pls 5-8.
2. BARY, A. DE 1887 Comparative Morphology and Biology of the Fungi, Mycetozoa and Bacteria. Eng. Trans. Clarendon Press, Oxford. Appeared in German, Leipzig, 1884.
3. 1886 Species der Saprolegnieen. Bot. Zeit. 48: 598-610, 614-21, 629-36, 545-53, Taf. 9 & 10.
4. BÜSGEN, ALEX. 1882 Die Entwicklung der Phycomycetensporangien. Jahr. f. wiss. Bot. 13: 253-285, 1 Taf.

5. CARLSON, M. C. 1929 Gametogenesis and Fertilization in *Achlya racemosa*. Ann. Bot. 43: 111-117, pl. 4.
6. CLAUSSEN, P. 1908 Über Entwicklung und Befruchtung bei *Saprolegnia monoica*. Festschr. Deutsch. Bot. Ges. 26: 144-161, pls. 6 and 7.
7. COKER, W. C. 1923 The Saprolegniaceae. Chapel Hill, North Carolina.
8. 1927 Other Water Molds from the Soil. Journ. Elish. Mit. Sci. Soc. 42: 207-226, pls. 27-36.
9. COKER, W. C., AND BRAXTON, H. H. 1926 New Water Molds from the Soil. Journ. Elish. Mit. Sci. Soc. 42: 139-149, pls. 10-15.
10. COKER, W. C., AND HYMAN, O. W. 1912 *Thraustotheca clavata*. Mycologia. 4: 87-90, 1 plate.
11. COOPER, G. O. 1929 A Cytological Study on Fertilization in *Achlya hypogyna* Coker and Pemberton. Trans. Wis. Acad. Sci. 24: 303-308, pl. 2.
12. Cytological Studies on the Sporangial Development and Gametogenesis in *Brevilegnia diclina* Harvey. Trans. Wis. Acad. Sci. 24: 309-322, pls. 3-5.
13. CORNU, MAX. 1872 Monographie des Saprolegniées. Ann. Sci. Nat., ser. 5, 15: 5-198, pls. 1-7.
14. COUCH, J. N. 1927 Some New Fungi from the Soil, with Observations on Spore Formation. Journ. Elisha Mitch. Sci. Soc. 42: 227-242, pls. 37-43.
15. 1932 The Development of the Sexual Organs in *Leptolegnia caudata*. Journ. Bot. 19: 584-599, pls. 42-44.
16. Unpublished Notes.
17. DAVIS, B. M. 1903 Oogenesis in *Saprolegnia*. Bot. Gaz. 35: 233-249, 320-349, pls. 9, 10.
18. 1905 Fertilization in the Saprolegniales. Bot. Gaz. 39: 61.
19. FISCHER, A. 1892 Saprolegniineae, in Rabenhorst's 'Kryptogamenflora'. 1 Tiel. Abt. 4, pp. 310-83, 13 figs., Ed. Kummer, Leipzig.
20. HARPER, R. A. 1905 Sexual Reproduction and the Organization of the Nucleus in Certain Mildews. Carnegie Inst. Washington Publ. 37: 104p., 7 plates.
21. HARTOG, M. M. 1895 On the Cytology of the Vegetative and Reproductive Organs of the Saprolegniaceae. Trans. Royal Irish Acad. 30: 649-708.
22. 1896 Fertilization in the Saprolegniales. Bot. Gaz. 39: 98.
23. 1899 The alleged Fertilization in the Saprolegniaceae. Ann. Bot. 13: 447.
24. HARVEY, J. V. 1925 A Study of the Water Molds and Pythiums Occurring in the Soils in Chapel Hill. Journ. Elish. Mit. Sci. Soc. 41: 151-164, pls. 12-19.
25. 1930 A Taxonomic and Morphologic Study of Some Members of the Saprolegniaceae. Journ. Elish. Mit. Sci. Soc. 45: 319-332, pls. 32-33.
26. HÖHNKE, VON W. 1935 Zur Cytologie der Oogon- und Entwicklung bei *Saprolegnia ferax* (Gruith) Thuret. Naturwissenschaftlichen Verein zu Bremen. 29: 308-323, 7 text figs.
27. HUMPHREY, J. E. 1893 The Saprolegniaceae of the United States. Trans. Am. Phil. Soc. 17: 63-148, pls. 14-20.
28. KASANOWSKY, V. 1911 *Aphanomyces laevis* de Bary, I. Entwicklung der Sexualorgane und Befruchtung. Ber. d. Deutsch. Bot. Ges. 29: 210-228, Taf. 10.

29. MÄCKEL, H. G. 1928 Zur Cytologie einiger Saprolegniaceen. *Jahr. wiss. Bot.* 69: 517-548, 25 text figs.
30. MINDEN, M. VON 1912 Phycomyceten, in *Kryptogamenflora der Mark Brandenburg* 5: 209-608.
31. MÜCKE, M. 1908 Zur Kenntnis der Eientwicklung und Befruchtung von *Achlya polyandra* de Bary. *Ber. d. Deutsch. Bot. Gesell.* 26: 367-378, pl. 6.
32. PATTERSON, P. 1927 Fertilization and Oogenesis in *Achlya colorata*. *Journ. Elisha Mit. Sci. Soc.* 43: 108-123, pls. 8-10.
33. FRINGSHEIM, N. 1859 Matériaux pour Servir à la Morphologie et à l'Étude Systematique des Algues. II Saprolegniées. *Ann. Sci. Nat., ser. 4*, 11: 349-371, pls. 6-7.
34. 1883 Nachträgliche Bemerkungen zu dem Befruchtungsact von *Achlya*. *Jahr. f. wiss. Bot.* 14: 111-131.
35. RAPER, J. R. 1936 Heterothallism and Sterility in *Achlya* and Observations on the Cytology of *Achlya bisexualis*. *Journ. Elisha Mit. Sci. Soc.* 52: 274-293, pls. 22, 23, 24.
36. SMITH, F. E. V. 1923 On the Direct Nuclear Divisions in Vegetative Mycelium of *Saprolegnia*. *Ann. Bot.* 37: 63-73, 12 text figs.
37. TROW, A. H. 1895 The Karyology of *Saprolegnia*. *Ann. Bot.* 9: 609-652, pls. 24, 25.
38. 1899 Observations on the Biology and Cytology of a New Variety of *Achlya americana*. *Ann. Bot.* 13: 131-179, pls. 8-10.
39. 1901 Observations on the Biology and Cytology of *Pythium ultimum* n. sp. *Ann. Bot.* 15: 289-311, pls. 15, 16.
40. 1904 On Fertilization in the Saprolegnieae. *Ann. Bot.* 18: 541-569, pls. 34-36.
41. 1905 Fertilization in the Saprolegniales. *Bot. Gaz.* 39: 300.
42. WESTON, W. H. 1918 The Development of *Thraustotheca clavata*, a Peculiar Water-Mould. *Ann. Bot.* 32: 155-173, pls. 4, 5.

EXPLANATION OF PLATES 12 AND 13

PLATE 12

- Fig. 1. Section of a young oogonium showing shape and appearance of nuclei in this stage in development. $\times 1135$. (a) Nuclei from a young oogonium enlarged. $\times 2015$.
- Fig. 2. Early prophase nuclei. $\times 2015$.
- Fig. 3. Late prophase nuclei showing the clustering of the chromatin into curious rod-like bodies. $\times 2015$.
- Fig. 4. Section of an oogonium showing (a) nuclei in metaphase of mitosis, (b) nuclei beginning to degenerate at this stage, and (c) granule-like remains of nuclei that began degeneration earlier. Nuclei in the antheridia are still in the resting stage. $\times 1135$.
- Fig. 5. Section of an antheridium showing mitosis. $\times 1135$. (See also section of antheridium in fig. 9.)
- Fig. 6. Enlarged metaphase nucleus. $\times 2015$.
- Fig. 7. Enlarged anaphase nucleus from an oogonium. $\times 2015$.

- Fig. 8. Later stage than shown in fig. 7 showing the chromatin becoming clustered together in the condition found in later stages in the development of the oogonium. $\times 2015$.
- Fig. 9. Section of an oogonium showing the female gamete nuclei. The other daughter nuclei are degenerating. Note mitosis taking place in a section of one antheridium. $\times 1135$.
- Fig. 10. Early stage in the cleavage of the egg origins. One gamete nucleus has not yet become oriented and no inward bulging of the cytoplasm has occurred. The other nucleus has its centrosome and rays in the typical position and the egg origin is forming. $\times 1135$.
- Fig. 11. Later stage in cleavage showing sections through three egg origins. $\times 1135$.

PLATE 13

- Fig. 12. Detail figure of an early stage in the formation of the egg origins showing the length and branching of the astral rays at this time. $\times 2015$.
- Fig. 13. Section of a large oogonium showing position of nucleus and acromatic figure during the rounding up of the oospheres. Some of the astral rays extend to the margin of the oosphere cytoplasm. $\times 1135$.
- Fig. 14. Smaller oogonium in same stage as that in fig. 13. Only the centrosome and rays show in one oosphere; the chromatin of the nucleus is in the adjacent section. $\times 1135$.
- Fig. 15. Oosphere after the membrane has formed. The astral rays have become much shorter. $\times 1135$.
- Fig. 16. Section of an oogonium showing two stages in fertilization. The fertilization tube at (a) is pushing against the oosphere membrane and the tip has become enlarged. At (b) the male nucleus has just been released and the end of the tube has collapsed somewhat. Note a "fertilization spot" in the last oosphere. $\times 1135$.
- Fig. 17. Section of an oogonium containing a single large egg showing the "penetration path" of the male nucleus. Astral rays are associated with both male and female nuclei at this time. $\times 1135$.
- Figs. 18-20. Stages in the development and fusion of the gamete nuclei. All figures except 20a $\times 1135$, 20a $\times 2015$.
- Fig. 21. Egg showing the zygote nucleus shortly after fertilization. $\times 1135$.
- Fig. 22. A longitudinal section of a sporangium showing astral rays and a centrosome associated with the nuclei in zoospores. $\times 1135$.

PLATE 12

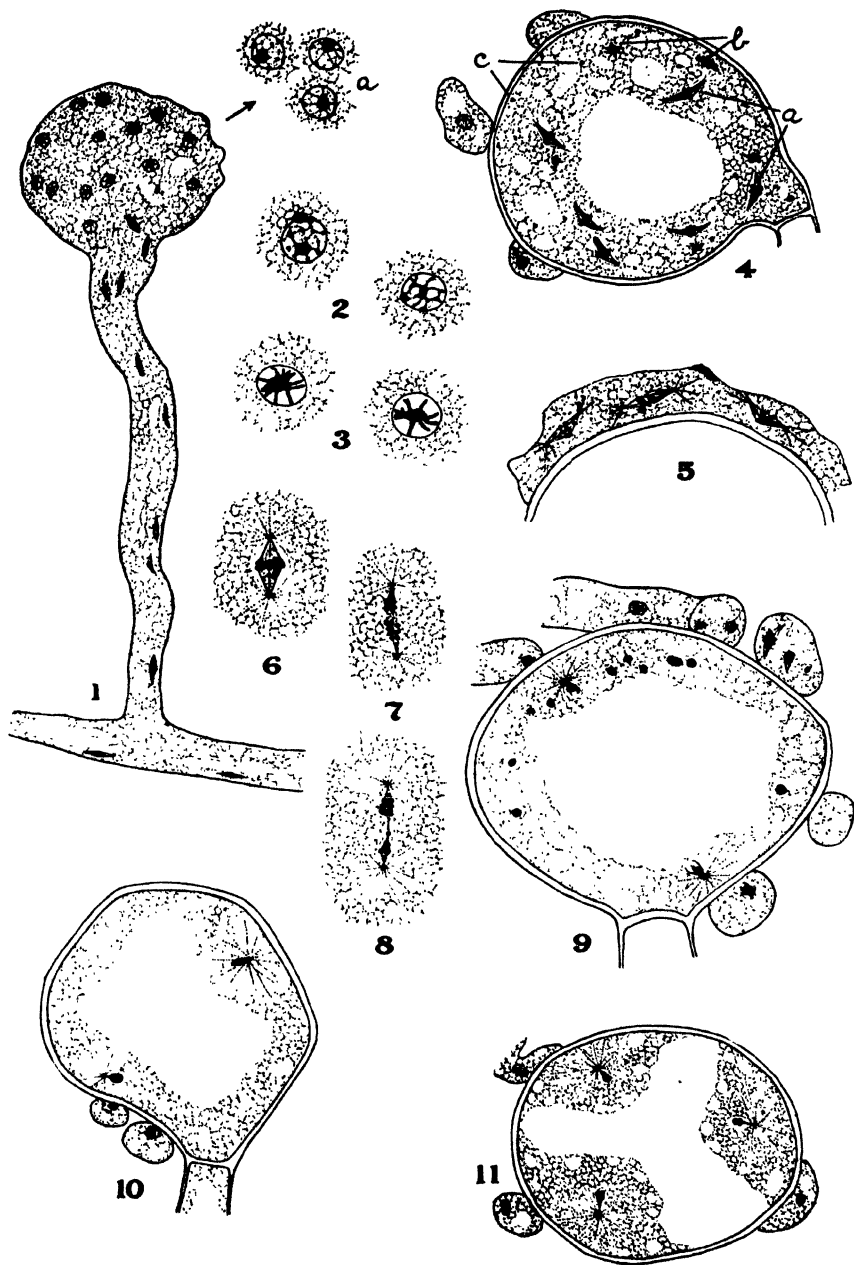
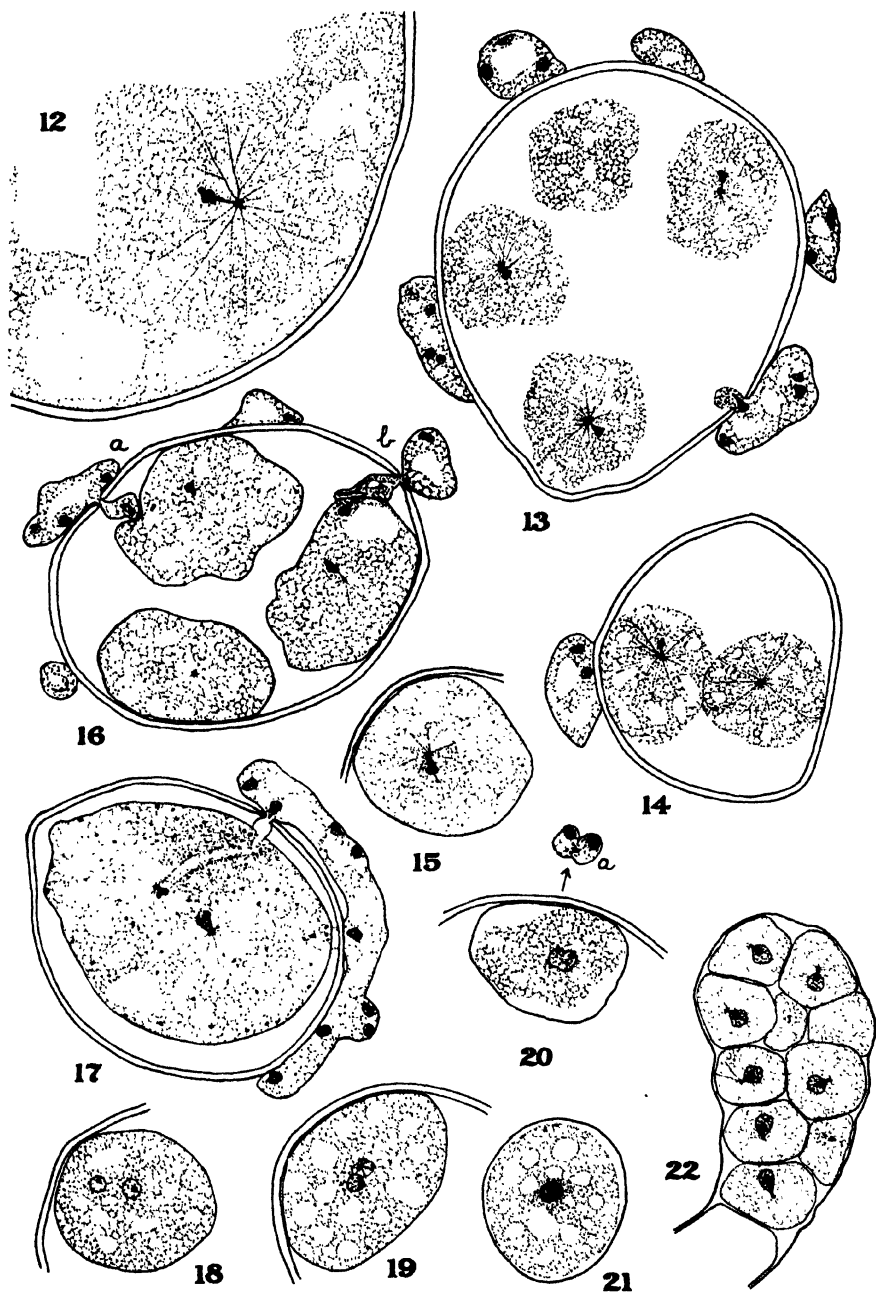


PLATE 13



THE OCCURRENCE OF *LIPARIS LOESELII* AND *HABENARIA BRACTEATA* IN NORTH CAROLINA

By LANE BARKSDALE

PLATE 14

In the *Manual of the Southeastern Flora* (1933), Small reports *Liparis Loeselii* as extending over an area covering "Ala. to Mo., Sask., Ont., and N.S." Though North Carolina falls into the range quoted above, no record of Loesel's twayblade had been made from this state before June, 1936.¹ For, though the author had known of the North Carolina colony prior to the above date (since June, 1931), he had made only photographic records of it, and the films constituting these records had been accidentally destroyed. Thus, in order to substantiate the reported existence of a colony of *L. Loeselii* (L.) L. C. Richard at Chimney Rock, N. C., it was necessary to make a return trip there in June, 1936.

About one half mile from Chimney Rock on Chimney Rock Mountain, Rutherford County, is Hickory Nut Falls, an exposed right-angled wall of granite, the crest of which rises a thousand feet above the fertile valley of the Rocky Broad River. Here to the left of the falls under a drip-rock, the colony of *Liparis Loeselii* grows in a mound of moss (a combination of *Sphagnum* and *Mnium*). Other inhabitants of the same mound include one or two *Vacciniums* (acting as a filter for the orchids against the morning sunlight), *Viola primulifolia*, one or two plants of *Habenaria clavellata*, and several grasses. The twayblade plants, the entire station numbering seven, sit with their pseudobulbs above ground, their strongly keeled leaves almost vertical, and their yellow-green spikes well hidden in the blades of grass. The number of flowers per spike ranges from two to fourteen.

Aside from the colony under the drip-rock, no other specimens of *L. Loeselii* have been found either on Chimney Rock Mountain or any other of the similar mountains bordering Hickory Nut Gorge, though quite a search has been made for them. Specimens of the twayblades

¹ Barksdale, Lane. Some Notes on the Orchids of Piedmont and Western North Carolina. Chapel Hill, N. C. 1936.

from the above station have been deposited in the Herbarium of the University of North Carolina, in the Gray Herbarium of Harvard, and in the Herbarium of Duke University.

So far as is known, *Habenaria bracteata* (Willd.) R. Brown has been collected heretofore in only one North Carolina county, and that collection was made by W. W. Ashe on Big Yellow Mountain, Mitchell County. On May 15, 1937, the author found one plant of *H. bracteata* growing in a clump of *Orchis spectabilis* near Cove Creek Post Office in Haywood County. Only one blossoming plant could be found. Nearby were a few seedlings growing in rich open soil, with large plants of *Trillium erectum* var. *album* and *Convallaria majalis*.

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PLATE 14



THE ORCHIDS OF NORTH CAROLINA

By DONOVAN S. CORRELL

PLATE 15

INTRODUCTION

This paper presents the results of a study of the orchid flora of North Carolina. The purpose of this study was to determine what species occur within the state, their habitats and distribution by counties and physiographical regions,* and the preparation of practical keys for their identification. No comprehensive study has been previously published of the orchids of North Carolina. However, many of the species found in the state have been included in various plant catalogues and manuals. In his unpublished "Flora Salemitana" (1821), which is in the possession of the Academy of Natural Sciences of Philadelphia, Schweinitz (19) listed 27 orchids as occurring in the district which includes Forsyth County and parts of several adjacent counties known as the "Salem Community." Curtis (1867) included 41 species of orchids in his botanical study of the state (13). In 1886, Hyams (15) listed along with other plants 2 species of orchids not included in Curtis' catalogue. The same year, in a list of the plants of Wilmington and vicinity, Wood and McCarthy listed 18 species of orchids, including the rare epiphyte *Epidendrum conopseum* R. Br. (23). Five years later (1891) Small and Heller (21), botanizing mainly in the western part of the state, collected 15 species of orchids. In 1936, Blomquist and Oosting (9) included 25 species of orchids in their spring and early summer flora of the piedmont.

Only 2 county lists, which include or concern orchids, have thus far been published for North Carolina. These are for Henderson and Guilford counties. In 1915, Memminger (16) published a list of plants growing "spontaneously" in Henderson County, which included 26 species of orchids. Most of these species have since been collected in Henderson County by Margaret C. Campbell and the writer. In 1933,

* This division follows that which is used by the United States Geological Survey; namely, the Coastal Plain Region, the Piedmont Region, and the Mountain Region.

Barksdale (7) published a paper on the orchids of Guilford County in which 17 species were discussed. Since then (1936) Barksdale (8) has published some notes on the orchids occurring in the piedmont and western North Carolina in which 38 species and 2 varieties were listed.

Besides the above publications the manuals of Chapman (11) and Small (20) include the flora of North Carolina. In the third edition (1897) of his manual, Chapman included 47 species in the Orchidaceae for North Carolina which have since then been definitely found to grow in the state. Small included North Carolina in the range of 51 species of orchids in his manual. Some of the species cited in the latter have not, however, been collected in the state, according to the specimens examined. Prof. Oakes Ames' revision of the Orchidaceae in Gray's *New Manual of Botany*, 7th edition (18), includes many of the orchids of the state.

The present study shows that 53 species and 5 varieties of orchids occur in North Carolina. Four of the species and 3 of the varieties have not been reported before from the state. In addition to the species which have been collected within the state, there are 3 others (*Microstylis spicata* Lindl., *Corallorrhiza trifida* Chatelain, and *Listera australis* Lindl.) which might be expected. The species not reported before from North Carolina are: *Listera cordata* (L.) R. Br., *Eulophia ecristata* (Fernald) Ames, *Spiranthes longilabris* Lindl., and *Spiranthes floridanum* (Wherry) and the forms *X Habenaria Canbyi* Ames, *X Spiranthes intermedia* Ames, and *Calopogon pulchellus* f. *albiflorus* (Britton) Fernald.

During the summer of 1935 a collecting trip, lasting from the middle of June until the middle of August, was made throughout North Carolina. Forty-five counties in the eastern half of the state and 32 counties in the western half were visited. In addition to these two extensive trips over the state, several others were made through the extreme southeastern counties and into counties adjacent to Durham County. About 500 collections were made on these trips. In addition to the collecting of specimens, field notes were taken concerning the different species found. The notes included such ecological data as were possible to be obtained from observation; habitat and plant associations were especially noted. Hydrogen-ion determinations were made with a Hellige Soil Tester on the soil in which many of the species grew, and flowers of most of the plants were preserved in 2% formalin for study in identification.

TAXONOMIC PROBLEMS

The most critical diagnostic characters of the Orchidaceae for practical purposes are the variations in the column and the size, shape, color, and position of the lip. Other characters which are valuable in some cases in separating the genera are the spur or nectary, as in *Habenaria*, and the type of inflorescence which is usually a solitary flower, a raceme, or a spike.

The use of vegetative parts of the plant in diagnosing and preparing keys for genera and species have been avoided as much as possible. In most cases they were found to be inconstant and therefore unreliable. Such is the case in *Spiranthes*, where the terms "leaves fugacious" and "leaves persistent" are much used in literature including the Orchidaceae. The leaves of those specimens of *Spiranthes* examined in this study, which included several thousand plants, were for the most part absent and were only in rare cases dependable as a diagnostic character. It seems permissible, however, to use the vegetative characters as secondary. Of course, in the case of caulescent and radical leaves, which are persistent throughout the season, as exemplified by the genus *Cypripedium*, it is possible to use the leaf character in distinguishing the species. For example, *Cypripedium acaule* Ait. with its two basal or radical leaves is strikingly different from *Cypripedium parviflorum* Salisb. with its leafy stem.

For several reasons, the genus *Spiranthes* may be considered to be the most difficult of the orchids found in North Carolina. Although several keys have been prepared for this genus (2, 18, 20), none seems to be adequate to cope satisfactorily with the difficulties encountered in this group. This may be readily understood from the fact that it is possible to take all the individual flowers on a spike in any single plant of the various species of *Spiranthes* and, upon careful examination, find some variation in the lip of every flower, although there will be a conformation to a certain type of lip which is outstanding in that particular spike. In spite of the variation of the lip in each plant or in different plants of the same species it seems, however, to be the most reliable character upon which to base a practical key. Such characters as the leaves, roots, glands, pubescence, bracts, and size of the plant in the genus *Spiranthes* must be used with discretion. Due to ecological variations, plants of the same species found growing in wet marshes and low meadows on the coast may be very different in size and appearance from those found in upland bogs and meadows in the piedmont and mountain region. Such variation must, therefore, be taken into

consideration in determining the species. The species of *Spiranthes*, as well as those of *Habenaria*, tend to hybridize freely (1, 4, 6, 14), thus causing additional confusion.

The closely allied species *Habenaria psycodes* (L.) Spreng. and *H. fimbriata* (Ait.) R. Br. are in many respects as difficult to separate as some species of *Spiranthes*. Ames says, "The difficulties are increased tenfold when it is realized that *Habenaria psycodes* and *H. fimbriata* are so similar that they are distinguishable only by arbitrary rules. In the preparation of the Orchidaceae for Gray's *New Manual* a conscientious effort was made to ascertain the distinctive characters of these two species. Although every conspicuous character was carefully studied it was found that the most reliable distinction was the depth of the fringe on the divisions of the labellum; in *H. psycodes* this being one-third the depth of the divisions or less, and in *H. fimbriata* one-third or more" (3). The writer finds that the size, number, and color of the flowers and the size of the spike are also helpful in distinguishing these two species: the flowers of *H. fimbriata* being larger, fewer, and paler than those of *H. psycodes*.

Some difficulty is encountered in the genus *Calopogon*. *Calopogon pulchellus* (Sw.) R. Br. may be separated from *C. parviflorus* Lindl. and *C. pallidus* Chapm. by its larger size, but the latter two species are often confused because the lip and column characters vary considerably in each. The middle lobe of *C. pallidus* is smaller than that of *C. parviflorus* but the shape may often be similar. *Calopogon parviflorus* blooms earlier, from April to early June, while *C. pallidus* blooms from late May until the last of July. All of the flowers in a raceme of *C. parviflorus* usually open about the same time while those of *C. pallidus* open successively from the base upward. The flowering period of the latter, therefore, usually extends over several weeks.

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PREFACE TO THE KEYS

The main objective in the preparation of the keys for the genera and species was to make them as practical and reliable as possible. Therefore, the keys are very descriptive and are based solely upon mature plants. The lip of the flower is the main diagnostic character used. Other flower characters have also been used in some cases, and these have been supplemented by vegetative characters where such were found to be distinctive enough to be of value.

An effort was made to make the keys as original as possible, but where confusion might arise by the introduction of new terminology, some of the diagnostic distinctions found in other keys were used. The manuals principally used for identification in the course of this work were Small's *Manual of the Southeastern Flora* (20) and Gray's *Manual* (18). The nomenclature used is based for the most part upon Prof. Oakes Ames' *Enumeration* (5), and the phylogenetic arrangement of the genera follows that of Pfitzer (17).

For completeness three species (*Microstylis spicata*, *Listera australis*, and *Corallorrhiza trifida*) which may be expected in the state but have

not thus far been found, are included in the keys. The distributional data for North Carolina which are cited for each species are based upon more than 2000 specimens examined. The general distributional data are compiled from various recent publications which include or concern the Orchidaceae (5, 10, 11, 18, 20).

In citing specimens the following abbreviations are used to designate the herbaria in which the specimens are deposited: (A)—Oakes Ames Herbarium, Harvard University, Cambridge, Mass.; (B)—Brooklyn Botanic Garden Herbarium, Brooklyn, N. Y.; (BU)—Brown University Herbarium, Providence, R. I.; (But.)—Butler University Herbarium, Indianapolis, Ind.; (C)—Cornell University Herbarium, Ithaca, N. Y.; (Da.)—May E. Day Herbarium, Oberlin, Ohio; (HMD)—H. M. Denslow Herbarium, Hartford, Conn.; (D)—Duke University Herbarium, Durham, N. C.; (MBD)—M. B. Dunkle Herbarium, Long Beach, Calif.; (F)—Field Columbian Museum, Chicago, Ill.; (G)—Gray Herbarium, Harvard University, Cambridge, Mass.; (GHG)—G. H. Grinnell Herbarium, Los Angeles, Calif.; (L)—Albert E. Lowmes Herbarium, Providence, R. I.; (NC)—University of North Carolina Herbarium, Chapel Hill, N. C.; (N)—New York Botanical Garden Herbarium, New York, N. Y.; (NS)—Academy of Natural Sciences Herbarium, Philadelphia, Pa.; (P)—University of Pennsylvania Herbarium, Philadelphia, Pa.; (S)—Salem College Herbarium, Winston-Salem, N. C.; (POS)—P. O. Schallert Herbarium, Winston-Salem, N. C.; (SC)—North Carolina State College Herbarium, Raleigh, N. C.; (T)—University of Tennessee Herbarium, Knoxville, Tenn.; (U)—United States National Herbarium, Washington, D. C.; (WV)—University of West Virginia Herbarium, Morgantown, W. Va.

KEY TO THE GENERA

1. Lip an inflated pouch; leaves large, plaited; fertile anthers 2
 1. *Cypripedium*
1. Lip concave or flat, not an inflated pouch; leaves various; fertile anther 1
 2. Flowers with a distinct and elongated spur, at least 2 mm. long
 3. Leaves present at time of flowering
 4. Flowers bicolorous; lip white, tongue-shaped, dilated; sepals and petals purple, uniting to form hood; leaves 1-2, basal; scape stout, 4-5 angled; flowering in early spring, April-May
 2. *Orchis*
 4. Flowers concolorous; lip variously fringed, lobed, notched, or divided; flowering later, middle June-August
 3. *Habenaria*
 3. Leaves absent at time of flowering, when present, green above, purplish beneath; flowers greenish, tinged with madder-purple; lip 3-lobed
 13. *Tipularia*

2. Flowers without a conspicuous spur; lip sometimes globose
5. Epiphytic; lip attached and adnate to the apex of the column, green, 3-lobed
14. *Epidendrum*
5. Terrestrial
6. Plants green; leaves 1-many, sometimes absent or inconspicuous at time of flowering in *Spiranthes* and *Aplectrum*
7. Lip crested or bearded on face
8. Leaves present at time of flowering; flowers white to purplish
9. Leaves linear to linear-lanceolate, more than 1, grass-like, sheathing the scape near the base; column winged at apex; lip above or superior, middle lobe prominent, basal lobes much reduced. 15. *Calopogon*
9. Leaves ovate to ovate-oblong, solitary near or above the middle of the stem, alternate along the stem, or in whorl of 5-6 at the top of the stem; column not winged, club-shaped; lip below or inferior
4. *Pogonia*
8. Leaves absent at time of flowering; flowers rose-purplish to pale purplish green or yellow
10. Flowers in raceme, pale purplish green or yellow; lip somewhat hastately 3-lobed, crested on the face with 3 longitudinal ridges; leaf solitary, large, oval, plaited, absent or withered at time of flowering 17. *Aplectrum*
10. Flower solitary (rarely 2), rose-purple; lip oblong, recurved, dilated, middle lobe pendent and eroded or fringed at apex, crested on face with 3-5 ranks of yellow or white fleshy hairs; leaf solitary, linear, developing after the flowering season 5. *Arthusa*
7. Lip not crested or bearded on face
11. Flowers essentially sessile, white or greenish white, in a compact spiral or cylindrical spike; sepals and petals, except lip, erect and connivent
12. Lip globose-saccate, with short, blunt tip, less than 6 mm. long, without basal callosities; raceme 1-sided or densely flowered; leaves ovate to ovate-oblong, in basal rosette, variegated with network of white veins; rooting from nodes of creeping rhizome
8. *Goodyera*
12. Lip not globose, but spreading, crisped and cut, concave, with callosities at base; flowers in 1-4 ranks, spirally twisted raceme; leaves never variegated, basal or extending up the stem, elliptic to linear-lanceolate; roots solitary or in a cluster at base of stem
6. *Spiranthes*
11. Flowers on pedicels at least 2 mm. long, variously colored, often minute, arranged in a loose raceme; sepals and petals free
13. Leaves basal, not cauline
14. Leaves numerous in basal rosette, spreading upon the ground; perianth greenish; lip small, concave, beaked, short-clawed, attached above the base to the column. 9. *Ponthieva*
14. Leaves not in basal rosette, although arising from the base, sheathing the stem
15. Leaves several, narrowly linear-elliptic, 20-50 cm. long, plicate,

- somewhat coriaceous; lip 3-lobed, as wide as long, 3 large lobes at base, rarely absent; plants slender, more than 35 cm. tall. 18. *Eulophia*
15. Leaves 1-2, ovate to elliptic-lanceolate, less than 20 cm. long, fleshy or membranous; plants small, less than 30 cm. tall
16. Lip less than 3.8 mm. long, cordate or auricled at the base, 3-lobed at the apex, broadest at the base, green or brownish green; flowers minute, less than 6 mm. long
10. *Microstylis*
16. Lip more than 3.8 mm. long, narrowed at the base, broadest and abruptly pointed at the apex, yellowish green or mauve; flowers larger, more than 6 mm. long. 11. *Liparis*
13. Leaves 2, not basal, cauline, near the middle of the stem, small round-ovate; lip cuneate or wedge-shaped, 2-lobed at the apex. 7. *Listera*
6. Plants not green; stem yellowish brown; leaves reduced to sheathing scales; rhizomes coralloid
17. Lip with 5-6 longitudinal ridges or crests down middle of face, 3-lobed; perianth about 2 cm. long. 16. *Hexaletris*
17. Lip not ridged or crested, at most lamellate, 3-lobed, entire, notched, or the margin denticulate, white or spotted with magenta-crimson; perianth less than 2 cm. long. 12. *Corallorrhiza*

1. CYPRIPEDIUM L. LADY SLIPPER, MOCCASIN FLOWER

1. Inflated pouch fissured in front; scape 1-flowered; lip usually pink, rose-veined on the face, with velvety appearance; leaves 2, basal and sheathing the stem, blades plaited. 1. *C. acaule*
1. Inflated pouch not fissured in front; peduncle 1- or more flowered; stem leafy, blades plaited
2. Lip longer than the sepals and petals, white to pale mauve, striped with white, flushed with rose-purple on face; sepals white. 2. *C. reginae*
2. Lip shorter than the sepals and petals, golden yellow, with glazed appearance; petals and sepals greenish, suffused with madder-purple
3. Lip small, 2-3 cm. long. 3. *C. parviflorum*
3. Lip large, 3.5-5 cm. long. 4. *C. pubescens*

1. *Cypripedium acaule* Ait. Pink moccasin flower (*Fissipes acaulis* (Ait.) Small)

Variable in habitat, in wet sphagnum bogs, low flood-plains along wooded streams, on rocky wooded slopes, and particularly where evergreens such as pines, hemlocks, and cedars are found, may be expected in any habitat where acid conditions prevail. April-July.

Mountain counties: Ashe: Leeds 1571 (NS). Buncombe: Biltmore 1211a (C, U), 1211b (N, U, P, G, B, F); Ramsey (NC); Pollard (U); Standley and Bollman 10029 (U); Correll 3840a (D). Burke: Alexander (NC); Blomquist and Correll 4702 (D). Henderson: Campbell (NC); Garren 229 (D); Correll 3269, 3342 (D). Macon: Landers (POS). Polk: Millsapugh (F); Day (Da.); Correll 3189 (D).

Transylvania: *House* 4190 (U); *Oosting* 36120 (D). Watauga: *Blomquist* 5927 (D); *Small and Heller* (F); *Schallert* (D); *Ashe* (NC); *Heller* (N).

Piedmont counties: *Burke*: *Correll* 3059, 3097 (D). *Davie*: *Blomquist and Correll* 4667 (D). *Durham*: *Blomquist* (P). *Forsyth*: *Penry* (S); *Denke* (S). *Granville*: *Correll* 469 (D, HMD), 589 (D). *Orange*: *Simmons* (NC). *Polk*: *Townsend* (U, C). *Wake*: *Correll* 308 (D).

Coastal Plain counties: *Edgecombe*: *Correll* 2446 (D). *Gates*: *Correll* 2276 (D). *Halifax*: *Correll* 2416 (D). *Johnston*: *Mitchell* (D); *Deans* (NC). *Martin*: *Fisher* (NS). *Perquimans*: *Glasson* (D).

Newfoundland and Nova Scotia to South Carolina and Alabama, west to Minnesota and Winnipeg.

2. *Cypripedium reginae* Walt. Queen lady slipper

(*C. spectabilis* Salisb.; *C. hirsutum* Mill.)

In swamps and on mossy wooded slopes. May-June.

Mountain counties: *Jackson*: *Ashe* (NC).

Newfoundland to Georgia, west to Missouri, Wisconsin, and Minnesota.

3. *Cypripedium parviflorum* Salisb. Small yellow lady slipper

On densely wooded banks or in floodplain habitats along streams, in neutral to acid soils. April-June.

Mountain counties: *Buncombe*: *Hogg* (N). *Burke*: *Coker* (NC). *Haywood*: *Blomquist* 5929 (D); *Standley* 5650 (U). *Henderson*: *Correll* 3248 (D, HMD), 3264 (D). *Macon*: *Landers* (POS); *Biltmore* 1208 (U, F, C), 1208b (B, F, U, G); —(G); *Correll* 3520 (D). *Mitchell*: *Wetherby* 190 (U). *Polk*: *Day* (Da). *Swain*: *Correll* 3702 (D). *Transylvania*: *Leeds* 1141 (NS); *House* 4149 (U), 4140 (U). *Watauga*: *Blomquist* 5928 (D); *Schallert* (S).

Piedmont counties: *Forsyth*: *Schweinitz* (NS). *Orange*: *Holland* (NC); *Deans* (NC). *Polk*: *Townsend* (U). *Randolph*: *Correll* 733, 736, 752 (D). *Wake*: *Blomquist and Correll* 303 (D).

Anticosti, Newfoundland, to Georgia and Mississippi, west through the Rockies to Washington and British Columbia.

4. *Cypripedium pubescens* Willd. Large yellow lady slipper

(*C. parviflorum* var. *pubescens* (Willd.) Knight)

On rich wooded slopes and in lowland thickets, neutral to mildly acid soils. April-June.

Mountain counties: *Buncombe*: *Biltmore* 157 (U, C); *Pollard* (U). *Burke*: *Biltmore* 157b (U). *Henderson*: *Campbell* (NC). *Macon*: *Harbison* (U); —(G). *Mitchell*: *Ashe* (NC); *Small and Heller* (F). *Transylvania*: *House* 4168 (U).

Piedmont counties: *Durham*: *Blomquist* 5930 (D). *Forsyth*: *Denke* (S). *Gaston*: *Correll and Blomquist* 5122 (D). *Orange*: *Smith* (NC); *Totten* (NC). *Polk*: *Townsend* (U, C). *Wake*: *Oosting* 3516A (D); *Blomquist and Correll* 302 (D).

Quebec and Nova Scotia to Georgia and Alabama, west to New Mexico and British Columbia.

2. ORCHIS L. ORCHIS

(Galeorchis Rydb.)

1. *Orchis spectabilis* L. Showy orchis

(Galeorchis spectabilis (L.) Rydb.)

Floodplain areas of streams in rich deciduous woods and in hemlock groves, neutral to slightly acid soils. May-July.

Mountain counties: Buncombe: *Biltmore* 679b (N, F, U, G, B), 679 (C); *Pollard* (U); *Standley and Bollman* 10070 (U); *Teague* (D); *Correll* 3841 (D, HMD). Haywood: *House* 5141 (N); *Standley* 5684 (U); *Price* 365 (D); *Oosting* 34252 (D); *Blomquist* 3952, 5953 (D). Henderson: *Holmes* (NC); *Campbell* (NC); *Correll* 3247 (D). Jackson: *Anderson* 1400 (P); *Oosting* 34330 (D). Mitchell: *Ash* (NC); *Wetherby* 191 (U). Polk: *Biltmore* 679c (N); *Day* (Da); *Peattie* 1633 (F, NC); *T wensend* 4 (C); *Correll* 3234 (D, A). Transylvania: *House* 4189 (U). Watauga: *Heller* (N); *Small and Heller* (C). Yancey: *Correll* 3874 (D).

Piedmont counties: Forsyth: *Schweinitz* (NS); *Lehman* (D); *Denke* (S); *Schal- lert* (S). Granville: *Correll and Blomquist* 5112 (D) Orange: *Totten* (NC); *Neely* (NC). Wake: *Oosting* 3517A (D).

New Brunswick and Ontario to Georgia, west to Nebraska, Missouri, and Arkansas.

3. HABENARIA WILLD. REIN ORCHIS, FRINGED ORCHIS

1. Lip deeply 3-parted, at least halfway to base
2. Divisions of the lip fringed or prominently crenate or eroded; petals not 2-parted, entire, crenate or fringed
3. Lateral lobes of lip capillary-fringed almost to base; middle lobe dilated at apex into a short-fringed wedge; petals entire; flowers pale yellowish or whitish green .. 1. *H. lacera*
3. Lateral lobes of lip fringed halfway to base at most, or eroded; flowers lilac, rarely white, to deep purple
4. Lobes of lip cuneate or flabellate, edges irregularly eroded, middle lobe notched at apex; petals slightly eroded or entire 2. *H. peramoena*
4. Lobes of lip broadly flabellate, copiously fringed; petals finely toothed
5. Lip more than 1.5 cm. broad, the lobes fringed to about one-third their depth; spike more than 4 cm. through, loosely flowered; flowers lilac 3. *H. ambriata*
5. Lip less than 1.5 cm. broad, the lobes fringed to less than one-third their depth; spike less than 3.5 cm. through, densely flowered and compact, flowers purple 4. *H. psychodes*
2. Divisions of the lip not fringed, entire; lateral lobes setaceous, much longer than the linear middle lobe; lip 8-9 mm. long; petals 2-parted nearly to the base; the lower segment setaceous, the upper linear and acute; flowers yellowish green in a loose raceme; spur as long as the ovary 5. *H. repens*
1. Lip not 3-parted, tongue-shaped, at most fringed, notched, or lobed

6. Lip copiously ciliate-fringed
7. Flowers white; lip narrowly ovate-lanceolate, 8-11 mm. long, coarsely fringed, basal fringes often branched.....6. *H. blephariglottis*
7. Flowers yellow or orange
8. Lip ovate, about 7 mm. long; spur shorter than the ovary, 5-9 mm. long
7. *H. cristata*
8. Lip oblong, about 1 cm. long; spur longer than the ovary, 1.5-2.5 cm. long
8. *H. ciliaris*
6. Lip not fringed, at most coarsely eroded
9. Lip entire or crenate, not lobed or notched
10. Lip tongue-shaped, ovate, entire or crenate on the margin, 4-4.5 mm. long; spur shorter than the ovary, 4-5 mm. long, awl-shaped; flowers yellow.....9. *H. integra*
10. Lip entire, ligulate or strap-shaped; flowers white or whitish green
11. Lip above, erect, narrowly oblong or lanceolate, 5-7 mm. long; spur twice as long as the ovary, ascending, about 1 cm. long; leaves mostly basal, linear, reduced on upper part of stem; flowers pure white
10. *H. nivea*
11. Lip below, oblong, obtuse, directed downward, 15-20 mm. long; spur longer than the lip, curved upward toward the tip, 2.5-4 cm. long; leaves orbicular, basal and radical; flowers whitish green
11. *H. orbiculata*
9. Lip hastately-lobed or notched at apex
12. Lip hastately 2-lobed at base, with horn-like tubercle on median line of face near base, apex truncate, rarely lobed, middle lobe 4.5-5 mm. long; spur slender, shorter than the ovary; flowers yellowish green
12. *H. flava*
12. Lip not hastately lobed at base, 3-lobed or toothed at apex; flowers greenish
13. Lip wedge-oblong, 3-5 mm. long, shallowly notched at apex into 3 short equal lobes; spur slender, clavate or 2-lobed, longer than the ovary; leaves 1-2, near middle of stem, oblong or spatulate
13. *H. clavellata*
13. Lip oblong-spatulate, 6-8 mm. long, unequally 3-lobed at apex, lateral lobes prolonged beyond obsolete middle lobe; spur saccate, very short, broad and blunt, whitish, delicate; floral bracts 2-4 times the length of the ovaries; stem leafy.....14. *H. bracteata*

1. *Habenaria lacera* (Michx.) Lodd. Ragged orchid
(*Blephariglottis lacera* (Michx.) Rydb.)

In moderately acid conditions in wet open sedge swamps and marshes, in meadows and glades of open woods. June-July.

Mountain counties: Ashe: Correll 4031 (D). Henderson: Campbell (NC); Biltmore 4815a (N, U). Macon: Blomquist 5944 (D). Mitchell: Wetherby 159 (U). Transylvania: House 3653 (G).

Piedmont counties: Forsyth: Schweinitz (NS); Denke (D). Union: Correll 921 (D, HMD, GHG, A).

Coastal Plain counties: Pender: *Wells* (SC).

Newfoundland to Georgia, west to Alabama, Missouri, and Manitoba.

2. *Habenaria peramoena* A. Gray. Purple fringeless orchid
(*Blephariglotis peramoena* (A. Gray) Rydb.)

In moderately to strongly acid soil of moist meadows and along stream banks. July–August.

Mountain counties: Buncombe: *Williamson* (NS); *Biltmore* 488b (N); *Hall* (B). Burke: *Correll* 3916 (D). Jackson: *Blomquist* 5947 (D). McDowell: *Hyams* (U). Macon: *Correll* 3494 (D). Mitchell: *Wetherby* (U). Swain: *Beardslee and Kofoid* (N, C); *Hall* (B). Watauga: *Carter* (P).

Piedmont counties: Caldwell: — (NC). Forsyth: *Shirley* (D); *Dente* (S). Iredell: *Hyams* (U).

Pennsylvania and New Jersey to North Carolina, west to Illinois, Missouri, and Alabama.

3. *Habenaria fimbriata* (Ait.) R. Br. Large purple fringed orchid
(*H. grandiflora* Torr.; *Blephariglotis grandiflora* (Bigel.) Rydb.)

In moist deciduous woods, in lowland meadows, medium to strongly acid conditions. June–August.

Mountain counties: Buncombe: *Biltmore* 4994 (P, U, N, NC, G), 4994b (N, U, G); *Standley and Bollman* 10003 (U). Haywood: *Oosting* 34455, 35390 (D); *Hall* (B). Mitchell: *Smith* (U); *Small and Heller* (U). Watauga: *Trudell* (HMD); *Durand* (C).

Newfoundland to North Carolina and Tennessee, west to Ontario and New York.

4. *Habenaria psycodes* (L.) Spreng. Small purple fringed orchid
(*Blephariglotis psycodes* (L.) Rydb.)

In upland meadows, swamps, and in open moist woods, neutral to slightly acid soil. June–August.

Mountain counties: Avery: *Ashe* 1337 (NC); *Small and Heller* (F). Buncombe: *Harvey* (NC); *Faxon* (G); *Correll* 57, 142, 3818, 3819 (D), 3839 (D, A). Haywood: *Blomquist* 5925 (D, P); *Smith* (U, G); *Wiegand* (C); *Standley* 5685 (U); *Hendrix* B65 62 (D); *Correll* 3788 (D, HMD). Jackson: *Blomquist* 5924 (D); *Williamson* (NS). McDowell: *Peattie* 939 (NC). Mitchell: *Small and Heller* (F, P); *Harshberger* 132 (P, N); *Ashe* (NC); *Schallert* 9437 (D); *Cannon* (U); *Wetherby* 160, 189 (U) *Smith* (U). Swain: *Anderson* 1465 (P); *Oosting* 35443 (D, HMD, GHG), 35420 (D); *Blomquist* 5940 (D); *Beardslee and Kofoid* (N, G, C). Transylvania: *House* 4227 (U). Watauga: *Trudell* (HMD); *Leeds* 1097 (NS); *Rydborg* 9501 (N).

Newfoundland to North Carolina, west to Minnesota and Iowa.

5. *Habenaria repens* Nutt. Creeping orchid

In ditches, streams, swamps, ponds, and on lake shores, often on floating mats on the surface of water. August–October.

Coastal Plain counties: Brunswick: *Reed* 98 (D). New Hanover: *Williamson* (NS); *Bartram* (NS); *Wherry* (NS); *Canby* 13 (NS, B, F); *Blomquist* 5945 (D).

Virginia to Florida, Alabama, and Louisiana; also West Indies, and Central and South America.

6. *Habenaria blephariglottis* (Willd.) Hook. White fringed orchid
(*Blephariglottis blephariglottis* (Willd.) Rydb.)

In strongly acid bogs, peat lands, and pine barrens. July–September.

Mountain counties: Haywood: *Standley* 5511 (U). Henderson: *Smith* 268 (BU, U); *Campbell* (NC); *Correll* 3353 (D, HMD), 3357 (D).

Coastal Plain counties: Bladen: *Heller* (F); *Blomquist* 5933 (D); *Blomquist and Correll* 2575 (D, GHG). Brunswick: *Blomquist* 5935 (P, D); *Matthews* (NC); *McCarthy* (U). Columbus: *Leeds* 1106 (NS); *Schallert* (POS, N, WV), 9405 (D). Craven: *Kearney* 1939, 1979 (U). Johnston: *Deans* (NC). Jones: *Alexander* (NC); *Beaven* 462 (D); *McCarthy* 84 (NC). Martin: *Fisher* (NS). Moore: *Katzenstein* (G). New Hanover: *Vestal* 36 (A); *Deans* (NC); *Williams* (U); *Smith* (U); *Ashe* 1010 (NC). Onslow: *Alexander* (NC); *Randolph* 994 (C, G). Pender: *Wells* (SC). Richmond: *Jester* (D, A). Wilson: *Blomquist* 7582 (D).

Newfoundland to Florida, west to Minnesota and Mississippi.

6a. *Habenaria blephariglottis* var. *holopetala* (Lindl.) A. Gray

Similar to the species in size, having narrower and obsolete toothed petals; lip entire or slightly serrate. In sphagnum bogs. July–September.

Mountain counties: Cherokee: *Correll* 3621 (D, HMD). Henderson: *Campbell* (NC).

Distribution same as the species.

6b. X *Habenaria canbyi* Ames

(*H. blephariglottis* x *H. cristata*)

Lip about 7 mm. long, deeply fringed; spur 12 mm. long; intermediate between the parent species in color and size of flowers (18). Acid bog. July.

Mountain counties: Henderson: *Correll* 3357 (D).

New Jersey, Delaware, and North Carolina.

7. *Habenaria cristata* (Michx.) R. Br. Crested orchid

(*Blephariglottis cristata* (Michx.) Raf.)

In decidedly acid habitats along the edge of open bogs and in low moist open woods. June–September.

Mountain counties: Henderson: *Campbell* (NC); *Holmes* (NC); *Smith* 158 (U, G); *Correll* 3361 (D, A, HMD).

Piedmont counties: Forsyth: *Schweinitz* (NS); *Denke* (S). Wake: *Ashe* (U).

Coastal Plain counties: Bladen: *Leeds* 1100 (NS); *Blomquist* 5938A (D); *Randolph* 1048 (C, G); *Heller* (F); *Blomquist and Correll* 2572 (D). Chowan: *Kearney* 1890 (U). Columbus: *Schallert* (POS, N), 5124 (D). Craven: *Biltmore* 4887a (P, U, G, N); *Brown* 16 (U); *Kearney* 1944, 1964 (U). Cumberland: *Leeds* 1099 (NS). Dare: *Abbe and Spalteholz* (C). Duplin: *Wherry and Trudell* (HMD). Edgecombe: *Alexander* (NC). Halifax: — (HMD). Johnston: *Deans* (NC); *Blomquist and Correll* 4770 (D). Jones: *Beaven* 460 (D, GHG). Lenoir: *Randolph* 789 (C). Martin: *Fisher* (NS). Moore: *Wicker* (NC); *Katsenstein* (G). Nash: *Blomquist* 7552 (D). New Hanover: *Wherry and Trudell* (HMD); *Williamson* (NS). Onslow: *Alexander* (NC). Pamlico: *Randolph* 888 (C). Pender: *Leeds* 1101 (NS). Richmond: *Smith* (D). Sampson: *Wherry and Trudell* (HMD). Wayne: *Burlingame* (BU). Wilson: *Blomquist* 7581 (D); *Randolph* 720 (C, G).

Massachusetts to Florida and Mississippi, west to Texas and Michigan.

8. *Habenaria ciliaris* (L.) R. Br. Yellow fringed orchid

(*Blephariglotis ciliaris* (L.) Rydb.)

In peaty bogs, meadows, edge of dense woods, floodplain regions, thickets, dry open soil. July–September.

Mountain counties: Ashe: *Ashe* (NC); *Sudworth* 107 (U). Avery: *Biltmore* 489b (P, G, U, NC, N, NS, F, B), 489 (C); *Gower* 529 (POS); *Rydborg* 9477 (N); *Standley and Bollman* 9974 (U); *Hall* (B); *Correll* 3951 (D). Burke: *Blomquist and Correll* 4708 (D); *Correll* 3901 (D, POS). Caldwell: *Seymour* 91 81 7 (C). Cherokee: *Correll* 3552 (D), 3605 (But, D), 3570 (D, HMD). Graham: *Correll* 3643 (D, GHG). Haywood: — (A); *Eliason* (NC); *Hall* (B). Henderson: *Smith* 270 (BU, U, G); *Campbell* (NC); *Correll* 3309, 3319 (D), 3335 (D, A). Jackson: *Correll* 3486 (D). Macon: *Beals* (HMD); *Coker* (NC); *Harbison* (G); *Harbison and Totten* (NC); *Correll* 3538 (D). Mitchell: *Ashe* (N); *Correll* 3885 (D, HMD, A, MBD). Polk: *Peattie* 1075 (NC). Swain: *Beardslee and Kofoid* (N, T, U, G, WV, B, F); *Correll* 3711 (D, GHG), 3760 (D). Transylvania: *Rydborg* 9500 (U, N, NS); *Smith* 269 (U); *Blomquist* 5936 (D); *Correll* 3370 (D). Watauga: *Carter* (P); *Sudworth* 47 (U); *Durand* (C); *Heller* (F). Yancey: *Leeds* 1186 (NS); *Abbe and Spalteholz* (C); *Correll* 3880 (D).

Piedmont counties: Forsyth: *Schweinitz* (NS); *Chitty* (D); *Denke* (S). Orange: *Totten* (NC); *Holland* 3422 (NC). Polk: *Townsend* (U, C). Richmond: *Jester* (D); *Smith* (D). Rowan: *Small* (F). Stanly: *Small and Heller* (N, F). Surry: *Schallert* 1016 (D). Wake: *Miller* (NC).

Coastal Plain counties: Bladen: *Randolph* 1045 (C, G); *Blomquist* 5938 (D). Brunswick: *Mattheus* (NC); *Munter* (G); *Blomquist* 5934, 5937 (D). Carteret: *Lewis* 97 (N). Columbus: *Schallert* (POS, N). Johnston: *Deans* (NC); *Ashe* (N, NC). Jones: *Beaven* 458 (D). Martin: *Fisher* (NS). Moore: *Wicker* (NC); *Katsenstein* (G). Nash: *Blomquist* 7553, 7568 (D). New Hanover: *Macfarlane and Nakahara* (P); *Vestal* 40 (A); *Williamson* (NS); *Williams* (U); *Smith* (U).

Onalow: *Ashe* (NC). Pender: *Blomquist* 5923 (D). Richmond: *Jester* (D); *Smith* (D). Scotland: *Smith* (D). Wayne: *Burlingame* (BU).

Ontario, Vermont, and Massachusetts, to Florida and Mississippi, west to Texas and Michigan.

9. *Habenaria integra* (Nutt.) Spreng. Yellow fringeless orchid
(*Gymnadeniopsis integra* (Nutt.) Rydb.)

In open moist pinelands and in open decidedly acid boglands of sphagnum or rotting wood. July–August.

Piedmont counties: Forsyth: *Schweinitz* (NS). Rowan: *Small* (F).

Coastal Plain counties: Brunswick: *McCarthy* (N). Martin: *Fisher* (NS).

New Jersey to Florida, west to Tennessee and Texas.

10. *Habenaria nivea* (Nutt.) Spreng. Snowy orchid
(*Gymnadeniopsis nivea* (Nutt.) Rydb.)

In swamps and low pinelands, and in wet open acid bogs. June–August.

Coastal Plain counties: Beaufort: *Correll* 1690 (D, A). Bladen: *Leeds* 1098 (NS); *Randolph* 1047 (C, G); *Blomquist and Correll* 2571 (D, HMD). Columbus: *Schallert* (POS); *Wiegand and Manning* 906 (C, G); *Heller* (F). New Hanover: *Trudell* (NS); *MacElwee* (NS); *Williamson* (NS); *Biltmore* 5202a (U, G). Pender: *McCarthy* (U); *Wells* (SC).

New Jersey and Delaware to Florida, west to Arkansas and Texas.

11. *Habenaria orbiculata* (Pursh.) Nutt. Large round-leaved orchid
(*Lysias orbiculata* (Pursh.) Rydb.)

In rich damp wooded ravines on mountain sides, acid habitats, July–August.

Mountain counties: Ashe: *Ashe* (NC). Mitchell: *Wetherby* 148 (N). Watauga: *Small and Heller* (P, N); *Huger* (N); *Gray* (N).

Piedmont counties: Stokes: *Denke* (D).

Labrador and Newfoundland to the mountains of South Carolina, west to Alaska, Washington, and British Columbia.

12. *Habenaria flava* (L.) R. Br. Tubercled orchid
(*Perularia flava* (L.) Farwell; *P. scutellata* (Nutt.) Small in part;
P. bidentata (Ell.) Small in part)

In wet grassy places and moist thickets in alluvial deposits, mountain bogs and low moist woods. June–September.

Mountain counties: Buncombe: *Biltmore* 1218 (U, C), 1218b (P, T, NS, N, U, NC, G, F). Cherokee: *Schweinitz* (NS). Haywood: *Blomquist* 5942, 5943 (D). Jackson: *Thaxter* (G). Macon: *Blomquist* 5941 (U).

Piedmont counties: Forsyth: *Schweinits* (NS); *Chitty* (D); *Denke* (D); *Lehman* (S).

Coastal Plain counties: Johnston: *Deans* (NC). Nash: *Oosting* 35578 (D). Wilson: *Wherry* (N).

Nova Scotia to Florida, along the Gulf Coast to Texas, westward to Minnesota, Illinois, and Missouri.

13. *Habenaria clavellata* (Michx.) Spreng. Little club-spur orchid
(*Gymnadeniopsis clavellata* (Michx.) Rydb.)

On the edge of streams in densely wooded areas, in sheltered sphagnum bogs, moist thickets, and swamps where moderately acid conditions exist. July–August.

Mountain counties: Ashe: *Correll* 3968 (D). Avery: *Correll* 3928 (D). Buncombe: *Peattie* 951 (NC); *Standley and Bollman* 10145 (U); *Biltmore* 490a (U, G); *Correll* 3831, 3848, 3860 (D). Burke: *Correll* 3913 (D). Cherokee: *Schweinits* (NS); *Correll* 3555 (D). Graham: *Correll* 3646 (D). Haywood: *Blomquist* 5939 (D); *Correll* 3791 (D). Henderson: *Wiegand and Manning* 896 (C, G); *Correll* 3251, 3362 (D). Jackson: *Ashe* (NC); *Blomquist* 5946 (D); *Thaxter* (G); *Correll* 3493 (D). Macon: *Coker and Holland* (NC); *Coker and Harbison et al* (NC); *Underwood et al* 2779 (T); *Buckley* (N). Mitchell: *Wetherby* (HMD); *Ashe* (NC); *Correll* 3888 (D, GHG). Polk: *Peattie* 978 (F), 1041 (NC). Swain: *Beardslee and Kofoid* (F, N); *Oosting* 35441 (D); *Correll* 3767 (D), 3713 (D, A, But, MBD, POS). Transylvania: *House* 3655 (G), 4376 (U); *Correll* 3391 (D), 3414 (D, HMD). Watauga: *Carter* (P); *Heller* (N, NS); *Robinson* 143 (G); *Hulst* (B).

Piedmont counties: Burke: *Correll* 3087 (D). Caldwell: *Randolph* 1125 (C, G). Caswell: *Ashe* (NC). Cleveland: *Correll* 3001 (D). Durham: *Correll* 4723 (D). Forsyth: *Schweinits* (NS); *Denke* (D, S); *Correll* 2652 (D). McDowell: *Correll* 3171 (D, HMD). Moore: *Blanknishop* 338 (A). Orange: *Alexander* (NC). Polk: *Townsend* (C, U). Warren: *Scholz* (NC).

Coastal Plain counties: Brunswick: *Blomquist and Correll* 4880 (D). Halifax: *Williamson* (NS). Harnett: *Blomquist and Correll* 2573 (D). Johnston: *Blomquist and Correll* 4768 (D). Nash: *Blomquist et al* 7577 (D). Pamlico: *Oosting* 33214 (D). Sampson: *Wherry and Trudell* (HMD). Wilson: *Oosting* 35585 (D).

Newfoundland to Florida, west to the Mississippi Valley, Minnesota, to Louisiana.

14. *Habenaria bracteata* (Willd.) R. Br. Bracted orchid
(*Coeoglossum bracteatum* (Willd.) Parl.)

Damp woods and thickets. May–June.

Mountain counties: Mitchell: *Ashe* (NC).

Piedmont counties: *Denke* (D).

Nova Scotia to North Carolina, west to Alaska, British Columbia, and Washington.

4. *POGONIA* JUSS. CREST LIP

1. Lip not lobed, at most lacerate-toothed, less than 2 cm. long; sepals and petals alike; flowers pale-rose to white; leaf 1, arising near middle of the stem, lance-ovate; a leafy bract at summit of stem. 1. *P. ophioglossoides*
1. Lip 3-lobed, slightly fimbriate along the margin
 2. Leaves 5-6 in a whorl at summit of stem, ovate, 2-5 cm. long, becoming larger as the erect fruit matures; sepals linear conduplicate, unlike the petals
 3. Sepals 5 cm. long; lip 2.5 cm. long, linear, partly papillose, with crest down middle; flower solitary, distinctly pedicelled. 2. *P. verticillata*
 3. Sepals about 2.5 cm. long, about the length of the petals, somewhat narrowed toward the base; lip crested over the entire face and on the middle of the lateral lobes; flower 1, often 2, essentially sessile. 3. *P. affinis*
 2. Leaves 1-several, not whorled, alternate
 4. Leaf solitary, rarely 2, arising near the middle of the stem, usually more than 2 cm. long; a leafy bract at summit of stem; flower 1, rarely 2; lip more than 2 cm. long. 4. *P. divaricata*
 4. Stem leafy; leaves wide-oval, concave, clasping the stem, less than 2 cm. long; flowers several, usually 3; lip less than 2 cm. long
5. *P. trianthophora*

1. *Pogonia ophioglossoides* (L.) Ker. Rose pogonia

In acid habitats, in wet open bogs, mossy tracts, and among sedges. May-June.

Mountain counties: Haywood: *Ruth* (T). Henderson: *Biltmore* 11028 (U); *Correll, Blomquist, and Garren* 5117 (D). Mitchell: *Ashe* (N).

Piedmont counties: Forsyth: *Schweinitz* (NS); *Lehman* (D); *Denke* (S); *Schallert* (S). Wake: *Coit* (SC).

Coastal Plain counties: Bladen: *Biltmore* 1369 (U, C); *Ashe* 347c (NC); *Blomquist and Correll* 399 (D), 400 (D, HMD, GHG, A). Brunswick: *Ashe* (NC); *Billings* (D); *Leeds* 1151 (NS). Columbus: *Schallert* (POS). Currituck: *Britton and Small* (N). Johnston: *Mitchell* (D). Martin: *Fisher* (NS). Moore: *Meritt* (L). New Hanover: *Ashe* (NC). Pamlico: *Correll* 1493 (D). Wayne: *Matthews* (NC). Wilson: *Blomquist* 7710 (D).

Newfoundland and New England to Florida and the Gulf States, west to Texas and Minnesota.

2. *Pogonia verticillata* (Willd.) Nutt. Large whorled pogonia

(*Isotria verticillata* (Willd.) Raf.)

In strongly acid soil on moist slopes in deciduous woods. April-July.

Mountain counties: Ashe: *Ashe* (NC). Buncombe: *Williamson* (NS); *Biltmore* 1246 (U, C), 1246b (N, U, G, F); *Martin* (C). Caldwell: *Small and Heller* (C, F). Haywood: *Blomquist* 5954 (D). Henderson: *Huger* (N); *Campbell* (NC); *Garren* 230 (D). Watauga: *Livingston* (N). Yancey: *Correll* 3870 (D, HMD, A).

Piedmont counties: Cleveland: *Correll* 3000 (D). Forsyth: *Lehman* (D, S).

Guilford: *Mitchell* (D). Polk: *Peattie* 1088 (NC, F). Surry: *Rusby* (N). Wake: *Kramer* (D).

Coastal Plain counties: Johnston: *Mitchell* (D).

New England to Florida and Louisiana, west to Texas, Arkansas, and Wisconsin.

3. *Pogonia affinis* C. F. Austin. Small whorled pogonia

On wooded slopes along stream banks. May-June.

Piedmont counties: Surry: *Douglas* (NC).

Erratically distributed in New England, New York, Pennsylvania, New Jersey, Maryland, Virginia, and North Carolina.

4. *Pogonia divaricata* (L.) R. Br. Spreading pogonia

(*Cleistes divaricata* (L.) Ames)

In strongly acid situations in swamps, meadows, and on wooded hillsides and mountain tops. May-July.

Mountain counties: Burke: *Gray et al* (NS, F). Henderson: *Wherry* (HMD); *Shoolbred* (N); *Correll* 3301B (D). Madison: *Oosting* 35332 (D); *Churchill* (T). Mitchell: *Wetherby* 61 (U). Polk: *Wheeler* (NS); *Millsbaugh* (F). Watauga: *Small and Heller* (C, U).

Piedmont counties: Burke: *Small and Heller* (P, HMD, N, NS, B, F). Caldwell: *Small and Heller* (F). Catawba: *Small and Heller* (F). Iredell: *Hyams* (U). Wake: *Lynn* (D).

Coastal Plain counties: Brunswick: *Blomquist and Correll* 444 (D); *Correll et al* 588 (D). Columbus: *Schallert* (POS). Craven: *Croom and Loomis* (N). Cumberland: *Schweinitz* (NS). Duplin: *Mathews* (NC). Harnett: *Correll* 2536 (D). Martin: *Fisher* (NS). New Hanover: *Burk* (P); *Bartram* (NS). Onslow: *Moltenke* 1246 (P, D, N).

New Jersey and Delaware, south to Florida and Alabama, west to Kentucky.

5. *Pogonia trianthophora* (Sw.) BSP Nodding pogonia

(*P. pendula* Lindl.; *Triphora trianthophora* (Sw.) Rydb.)

In damp rich woods, thickets, or in evergreen forests in neutral to slightly acid soil. July-September.

Mountain counties: Buncombe: *Beals* (A, HMD); *Correll* 3845 (D, POS). Burke: *Alexander* (NC). Haywood: *Smith* (F, NS, U, G); *Coker* (NC); *Ruth* 430 (G). Henderson: *Memminger* (N); *Campbell* (NC); *Holmes* (NC); *Wiegand and Manning* 920 (C). Jackson: *Williamson* (N, NS). McDowell: *Leads* 1148 (NS). Macon: *Coker et al* (NC). Mitchell: *Wetherby* (HMD). Polk: *Peattie* 1511 (NC). Swain: *Beardslee and Kofoid* (A, N, G, WV, F); *Hulst* (B). Transylvania: *Emory* (A); *Wherry* (HMD); *Slosson* (G); *Correll* 3409, 3425 (D), 3434 (D, A, GHG, HMD). Watauga: *Durand* (C); *Yancey*: *Biltmore* 781b (N, U, G); *Hall* (B).

Piedmont counties: Forsyth: *Denke* (D); *Schuman* (S). Polk: *Townsend* (U, C).

Maine to Florida and Alabama, west to Wisconsin and Missouri.

5. ARETHUSA (GRONOV.) L. ARETHUSA

1. *Arethusa bulbosa* L. Bog arethusa
In acid sphagnum bogs. May-June.

Mountain counties: Henderson: *Campbell* (NC); *Shoalbred* (N). Transylvania: *Huger* (N).

Piedmont counties: Forsyth: *Schweinitz* (NS).

Newfoundland to mountains of South Carolina, west to Minnesota and Wisconsin.

6. SPIRANTHES RICH. LADIES' TRESSES

(*Ibidium* Salisb.; *Gyrostachys* Pers.)

The genus may be distinguished by the arrangement of the small, white, yellowish or greenish white flowers in a more or less spirally twisted raceme, often merely secund; lateral lanceolate sepals free to the base, not decurrent on the ovary; upper sepal united with the oblong petals; lip short-stalked, with a callus protuberant (callosity) within on each side of the base.

According to Ames (4), "*Spiranthes* is the most perplexing orchid genus in our flora. It is the least understood and the one that furnishes to authors who grow impatient under the restraints imposed by cautious progress, the best opportunities for the multiplication of species. It is a genus that repays intensive observation in the field and prolonged contemplation in the herbarium."

1. Lip less than 5 mm. long
2. Raceme densely flowered in several ranks, not conspicuously twisted, oval in appearance; lip ovate, sometimes constricted above the middle, about 4 mm. long; callosities slender, elongated, strongly curved; leaves persistent, radical and caudicle, linear, acute, less than 10 cm. long; flowers small, white, nodding..... 1. *S. ovalis*
2. Raceme loosely flowered, in single rank, spirally twisted, often secund; leaves fugacious, when present basal, small, ovate, elliptic
3. Lip white, quadrate, flared at apex, crisped, 2-3 mm. long; root solitary (the remains of last year's tuber often persisting)..... 2. *S. Beckii*
3. Lip medially colored, 3-4 mm. long; roots fasciculate
4. Lip medially yellow, ovate to ovate-oblong, slightly constricted above the middle, flared and crisped at the apex; callosities slender; flowers opening in spring, April-May..... 3. *S. floridanum*
4. Lip medially green, oblong-quadrate, with a crisped white margin; callosities stout; flowers opening in summer and fall, June-September
4. *S. gracilis*
1. Lip 5 mm. long or more
5. Raceme loosely flowered in single rank, merely secund or spirally twisted; leaves persistent or fugacious

6. Lip 8-9 mm. long, broadly rhombic at base, 4-5 mm. wide, tapering to the obtuse tip, wavy on the margin, crisped above base, recurved near apex; flowers secund in a single straight or scarcely spiral row, horizontal; bracts ovate, acute, scarcely longer than the ovaries, smooth, glabrous; basal leaves wanting during anthesis; flowering in the fall, September-November.....5. *S. longilabris*
6. Lip 5-8 mm. long; flowering in the spring or early summer, April-July
7. Lip oblong, often flared and broadest at the distal end, smooth beneath, thinnish, the veins prominent, 6-8 mm. long; flowers white, often veined with green; leaves mostly fugacious.....6. *S. praecox*
7. Lip ovate to ovate-oblong, broadest in front of the callosities, pubescent beneath, more opaque and thicker, with nerves showing less distinctly than in the preceding species, 5-7 mm. long; linear-lanceolate leaves mostly persistent.....7. *S. vernalis*
5. Raceme densely flowered in 2-several ranks (rarely 1-ranked in *S. cernua*), the rachis not conspicuously twisted; leaves persistent
8. Lip 9-11 mm. long, basal half dilated, rhomboidal, tapering to the obtuse apex, often broadly ovate to broadly cuneate, callosities prominent; flowers cream-colored, often marked with green, fragrant; leaves extending up the stem.....8. *S. odorata*
8. Lip 7-10 mm. long, ovate-oblong, usually constricted about the middle and flared at the apex; flowers white, slightly downy, nodding perceptibly; leaves mostly radical.....9. *S. cernua*

1. *Spiranthes ovalis* Lindl. Oval ladies' tresses Plate 15, f. 1
(*Ibidium ovale* (Lindl.) House)

In moist shady woods and on the edge of thickets, on wooded hills. September-October.

Piedmont counties: Durham: *Correll* 4751, 4754 (D). Orange: *Barksdale* (NC).

North Carolina to Georgia, westward to Texas, up the Mississippi Valley to Illinois.

2. *Spiranthes Beckii* Lindl. Little ladies' tresses. Plate 15, f. 2
(*Gyrostachys simplex* (A. Gray) Kuntze; *Ibidium Beckii* (Lindl.) House)

In dry soil in fields and open woods, acid habitats. June-September.

Mountain counties: Cherokee: *Correll* 3553B (D). Graham: *Correll* 3640A (D). Swain: *Beardslee and Kofoed* (N, C, F).

Piedmont counties: Durham: *Latham* (D); *Correll* 4656, 4755A (D), 4656A (D, A). Forsyth: *Schallert* 1300 (D); *Correll* (D). Iredell: *Blomquist and Correll* 4670 (D). McDowell: *Blomquist and Correll* 4672 (D, HMD, GHG). Orange: *Reasoner* (NC); *Alexander* (NC); *Ashe* 553 (NC); *Correll* 4725 (D), 4742 (D, MBD). Randolph: *Barksdale* (NC). Rowan: *Small and Heller* (F). Stokes: *Schallert* 8136 (POS).

Coastal Plain counties: Beaufort: *Correll* 1561A, 1704A (D). Camden: *Correll* 2216A (D). New Hanover: *Williamson* (NS). Pamlico: *Correll* 1492A, 1502A (D). Richmond: *Wiegand and Manning* 911 (C, G). Sampson: *Wherry and Trudell* (HMD). Tyrrell: *Correll* 1895A (D). Washington: *Correll* 1901A (D, POS).

Massachusetts to Florida and Texas, up the Mississippi Valley to Arkansas and Kentucky.

3. *Spiranthes floridanum* (Wherry) comb. nov. Florida ladies' tresses (*Ibidium floridanum* Wherry) Plate 15, f. 3.

Savannah land. May.

Coastal Plain counties: Pender: *Wells* (D, SC).

Coastal Plain, North Carolina, to Florida and east Texas.

4. *Spiranthes gracilis* (Bigel.) Beck Slender ladies' tresses (*Ibidium gracile* (Bigel.) House) Plate 15, f. 4

In dry, moderately acid soil, grassy fields, open woods, and on beach lands along the coast. June–September.

Mountain counties: Ashe: *Correll* 4035 (D). Burke: *Alexander* (NC). Caldwell: *Small and Heller* (F). Cherokee: *Correll* 3622, 3637 (D), 3553 (D, POS). Graham: *Correll* 3640, 3682 (D). Haywood: *Standley* 5739 (U); *Hall* (B); *Hall* 509 (U). Henderson: *Holmes* (NC); *Correll* 3340 (D). Jackson: *Correll* 3488, 3488A (D). McDowell: *Standley and Bollman* 10110 (U). Mitchell: *Wetherby* (HMD). Polk: *Peattie* 1109½ (NC). Swain: *Beardslee and Kofoed* (N, C); *Wright et al.* 234 (C). Transylvania: *House* 4015 (G). Watauga: *Durand* (C).

Piedmont counties: Caldwell: *Randolph* 1064 (C). Durham: *Latham* (D); *Correll* 4657, 4753 (D). Forsyth: *Schweinitz* (NS); *Lehman* (S); *Correll* 4094 (D). Orange: *Reasoner* (NC). Polk: *Townsend* (C, U). Randolph: *Barksdale* (NC). Rowan: *Heller* (NS); *Small and Heller* (F). Warren: *Scholz* (NC).

Coastal Plain counties: Beaufort: *Brues* (A); *Correll* 1704 (D, A, MBD), 1561, 1579 (D). Bertie: *Correll* 1958 (D). Camden: *Correll* 2216, 2217 (D). Chowan: *Wiegand and Manning* 913 (G); *Correll* 2018 (D). Cumberland: *Schallert* (POS). Currituck: *Correll* 2616 (D). Johnston: *Smith* (NC). Pamlico: *Correll* 1492, 1502 (D). Pasquotank: *Correll* 2085 (D). Tyrrell: *Correll* 1895 (D, A, HMD, GHG). Washington: *Correll* 1901 (D, But).

Nova Scotia to Georgia and Louisiana, west to Texas and Manitoba.

5. *Spiranthes longilabris* Lindl. Long-lipped ladies' tresses (*Ibidium longilabre* (Lindl.) House) Plate 15, f. 5

Open savannah land in the coastal plain. October.

Coastal Plain counties: Pender: *Wells* (SC, D).

North Carolina to Florida, Mississippi, and Louisiana.

6. *Spiranthes praecox* (Walt.) S. Wats. and Coult. Giant ladies' tresses

(*Ibidium praecox* (Walt.) House; *Gyrostachys praecox* (Walt.) Kuntze)
Plate 15, f. 6

In wet grassy bogs and low moist meadows and marshes. May-August.

Piedmont counties: Anson: *Correll* 1040 (D). Person: *Oosting* 33335A (D).

Coastal Plain counties: Beaufort: *Correll* 1560, 1676A, 1696A, 1705B (D). Bladen: *Correll* 402 (D). Brunswick: *Leeds* 1122 (NS); *Correll* 445A, 587A, 456 (D). Camden: *Correll* 2065, 2218, 2219 (D). Carteret: *Lewis* 96 (N). Columbus: *Schallert* (D, POS). Craven: *Correll* 1473A (D, POS). Currituck: *Correll* 2131, 2150, 2138A, 2139 (D). Dare: *Correll* 1738 (D, HMD, A, GHG). Greene: *Correll* 1342 (D, A). Martin: *Correll* 1844A (D). Nash: *Correll* 2500 (D). New Hanover: *Coville* (U); *Macfarlane* (P); *Williamson* (NS). Pamlico: *Correll* 1489, 1491, 1496 (D). Pender: *Wells* (SC). Richmond: *Wherry* (NS); *Correll* 1111 (D). Scotland: *Correll* 1179 (D). Washington: *Correll* 1902 (D).

New Jersey to Florida and Alabama, westward to Texas.

7. *Spiranthes vernalis* Eng. and Gray Spring ladies' tresses

(*Ibidium vernale* (Eng. and Gray) House) Plate 15, f. 7

In low pastures and meadows, bogs, marshes, swamps, and in open dry gravelly soil. May-August.

Mountain counties: Ashe: *Correll* 4036 (D). Cherokee: *Correll* 3553A (D).

Piedmont counties: Anson: *Correll* 1029 (D, POS). Cabarrus: *Correll* 851 (D). Durham: *Blomquist* 7401 (D); *Oosting* 33132 (D). Granville: *McCarthy* (U). Orange: *Coker and Totten* (NC). Rowan: *Heller* (N). Stanly: *Correll* 918 (D). Union: *Correll* 956, 990 (D), 999 (D, But.), 999A (D, GHG), 920 (D, HMD, A). Wake: *Blomquist* 3533 (D).

Coastal Plain counties: Beaufort: *Correll* 1675, 1705, 1705A, 1696, 1706, 1706A, 1627 (D), 1676 (D, MBD). Bertie: *Correll* 1953, 1984 (D). Brunswick: *Wiegand and Manning* 915 (G); *Correll* 455 (D, HMD), 587, 2132A (D). Camden: *Correll* 2065A (D). Chowan: *Wiegand and Manning* 913 (C); *Correll* 2019, 2020 (D). Craven: *Correll* 1473 (D), 1474 (D, A). Currituck: *Blomquist* (D); *Correll* 2130, 2137, 2181, 2131A, 2138, 2136, 2155 (D). Dare: *Blomquist* 7453 (D). Edgecombe: *Correll* 2434 (D). Gates: *Correll* 2241 (D). Greene: *Correll* 1341 (D). Halifax: *Williamson* (NS). Martin: *Correll* 1844 (D). New Hanover: *Wells* (SC). Pamlico: *Correll* 1500, 1501, 1501A (D). Tyrrell: *Correll* 1894 (D).

Massachusetts to Florida, west to Mexico, Illinois, and Missouri.

7a. X *Spiranthes intermedia* Ames

(*S. gracilis* x *S. vernalis*)

Resembling *S. vernalis* but more slender throughout, 18-30 cm. tall; spike 3-7 cm. long; lip 4.5 mm. long, elliptic-oblong, greenish with green, slender callosities, wavy-margined at distal end; floral bracts 5 mm.

long, ovate-lanceolate, strongly hyaline-margined, somewhat translucent; leaves mostly basal, linear-lanceolate, about 5 cm. long. In upland meadows and in dry meadows along the coast. July.

Coastal Plain counties: Currituck: *Correll* 2138A, 2132 (D).
Massachusetts and North Carolina.

8. *Spiranthes odorata* (Nutt.) Lindl. Fragrant ladies' tresses
(*Gyrostachys odorata* (Nutt.) Kuntze) Plate 15, f. 8

In low marshes and swamps, often growing in the water of flooded swamps in dense clumps because of its stoloniferous habit. October–November.

Coastal Plain counties: Brunswick: *Blomquist and Correll* 4893 (D). Columbus: *Reed* 24, 121 (D); *Blomquist and Correll* 4897 (D, HMD, A, GHG). Craven: *Loomy* (N); *McCarthy* (NS). Currituck: *McAtee* 2933 (N). Halifax: *Bartram* (NS).

Maryland and Virginia and along Gulf States to Texas.

9. *Spiranthes cernua* (L.) Rich. Nodding ladies' tresses
(*Ibidium cernuum* (L.) House) Plate 15, f. 9

In strongly acid soils in bogs, swamps, low meadows, and on the edge of low swampy woods. September–November.

Mountain counties: Ashe: *Ashe* (NC); *Eggleston* 4177 (U). Buncombe: *Biltmore* 3517a (U, P, G, N); *Ashe* (NC). Haywood: *Huger* (N). Henderson: *Campbell* (NC). Jackson: *Wherry and Pennell* 14199 (NS). Yancey: *Beals* (HMD); *Wherry and Pennell* 14269 (NS).

Piedmont counties: Durham: *Reed* 12 (D); *Blomquist* 5955 (D); *Blomquist and Correll* 4901 (D); *Duncan* (D); *Correll* 4913, 4909 (D). Forsyth: *Denke* (D, S); *Lehman* (D). Franklin: *Oosting* 34802 (D, HMD, GHG). Granville: *Reed* (D). Orange: *Coker* (NC). Randolph: *Causey* (NC); *Barksdale* (NC). Wake: *Burlingame* (BU).

Coastal Plain counties: Brunswick: *Wiegand and Manning* 915 (C). Hertford: *Wherry* (P). Hoke: *Schallert* (POS). Johnston: *Blomquist* (D). Moore: *Blomquist* 7641, 7642 (D, A.); *Holland* (NC); *Meritte* (L.). New Hanover: *Schallert* (D); *Bartram* (NS).

Newfoundland to Florida, west to Texas, New Mexico, up the Mississippi Valley to Minnesota and South Dakota.

9a. *Spiranthes cernua* var. *ochroleuca* (Rydb.) Ames

To differentiate the variety from the species, Ames says: "There is only one sure guide that I have found satisfactory, namely, polyembryonic seeds for the species and normal seeds for the variety" (4). The variety grows in drier ground than the species, blooms somewhat later, and has longer floral bracts. The flowers are greenish, cream-

colored, or white. In open dry forests or edge of woods. September–November.

Piedmont counties: Randolph: *Causey* (NC).

Maine, South Dakota to Georgia, and New Mexico.

7. LISTERA R. BR. TWAYBLADE

(*Ophrys* L.)

1. Lip with a tooth on each side at base
2. Lip wide-ovate to round-cuneate, 7–9 mm. long, 7–10 mm. across apex, cleft at apex, toothed in the fork of the lobes; basal lateral teeth curved, oblong; leaves ovate-reniform or kidney-shaped, mucronate; flowers whitish, long-pedicelled; pedicels over 6 mm. long.....1. *L. Smallii*
2. Lip narrowly oblong, 3–4 mm. long, cleft halfway to base, lobes lance-linear; basal lateral teeth curved, horn-like; leaves round-ovate, somewhat heart-shaped; flowers purplish to yellowish green, short-pedicelled; pedicels less than 4 mm. long.....2. *L. cordata*
1. Lip without basal teeth, lance-linear, 6–12 mm. long, cleft halfway to base, lobes linear-setaceous, acute; leaves wide-ovate; flowers reddish purple, slender-pedicelled; pedicels over 7 mm. long.....3. *L. australis*

1. *Listera Smallii* Wiegand Small's twayblade, Kidney-leaved twayblade

(*Listera reniformis* Small; *Ophrys Smallii* (Wiegand) House)

On moist wooded mountain slopes, or in damp shaded rhododendron thickets, in strongly acid habitats and often in wooded sphagnum bogs. June–July.

Mountain counties: Alleghany: *Correll* 4072 (D). Avery: *Trudell* (NS); *Leeds* 1615 (NS); *Correll* 3943 (D). Buncombe: *Harshberger* 90 (P, NS); *Standley and Bollman* 10030 (U); *Martin* (C); *Frank* (N); *Correll* 3849 (D), 3832 (HMD, D, POS). Burke: *Schum* (P); *Curtis* (N); *Gray et al.* (NS). Haywood: *Correll* 3789 (D). Jackson: *Huger* (HMD, N, D); *Blomquist* 5950 (D); *Trentham* (D); *Ashe* (NC). McDowell: *LeRoy and Ruger* (HMD); *Peattie* 954 (NC). Macon: *Coker and Holland* (NC); *Boynnton* (U); *Wright* (C). Mitchell: *Price* (NS); *Merriam* (U); *Wetherby* 65 (U); *Wetherby* (HMD); *Ashe* (NC); *Gray* (G). Swain: *Beardslee and Kofoid* (F, N). Transylvania: *House* 3671 (G). Watauga: *Small and Heller* (P, BU, A, NS, F, U, N, C); *Trudell* (L, HMD, BU); *Wherry* (U); *Sudworth* (U); *Randolph* 1182 (G, C). Yancey: *Rydberg* 9385 (N, U); *Leeds* 1196 (NS); *Holmes* (NC); *Correll* 3876 (D, GHG, A, But.).

Pennsylvania to North Carolina in the mountains.

2. *Listera cordata* (L.) R. Br. Heart-leaved twayblade

High mountain regions, or mossy damp forest floors, particularly under evergreens, also in sphagnum bogs. June–July.

Collections: "North Carolina": *Chapman* (U). "Collected in the mountains of North Carolina, foot of Black, June, 1872": *LeRoy and Ruger* (F). "In the mountains of Virginia and North Carolina, July, 1841": *Gray and Carey* (N).

Newfoundland and Labrador to North Carolina, westward to the Pacific Coast, from Alaska to New Mexico.

3. *Listera australis* Lindl. Southern twayblade

Although North Carolina is within the range of this species, no specimens have been seen from the state. Curtis (13) includes it in his catalogue, and Wiegand cites the species in his revision of the genus (22). Wood and McCarthy (23) reported it for New Hanover County, "July. Flowers small, greenish." April-July.

Ontario and Vermont to Florida and Louisiana, along the coastal plain region.

8. GOODYERA R. BR. RATTLESNAKE PLANTAIN

(*Epipactis* (Haller) Boehmer; *Peramium* Salisb.)

Raceme loosely few-flowered, 1-sided, about 4.5 cm. long; lip saccate with an elongated tip, margin flared or recurved; flowers white tinged with green; leaves dark green; veins of leaf conspicuously bordered with white, confluent

1. *G. repens* var. *ophioides*

Raceme densely many-flowered, cylindrical, about 7 cm. long; lip strongly saccate with short blunt tip, margin not flared or recurved; flowers white; leaves bluish green; veins finely bounded with white, not confluent

2. *G. pubescens*

1. *Goodyera repens* var. *ophioides* Fernald Lesser rattlesnake plantain (*Peramium repens* (L.) Salisb.; *Peramium ophioides* (Fernald) Rydb.; *Epipactis repens* var. *ophioides* (Fernald) A. A. Eaton)

Trailing, stoloniferous, in dense mats of moss in damp cold woods, chiefly beneath evergreens, moderately acid habitat. June-August.

Mountain counties: Avery: *Seymour* 9188 (C, G). Buncombe: *Correll* 3835 (D, HMD, GHG, A). Haywood: *Anderson* 1449 (P); *House* 4490 (U). Madison: *Ashe* (NC). Swain: *Beardslee and Kofoid* (G). Watauga: *Small and Heller* (P, NS, U, F); *Heller* (N, NS); *Durand* (C). Yancey: *Billmore* 5199a (N).

Newfoundland and Nova Scotia to South Carolina, west to Manitoba, Michigan, and New Mexico.

2. *Goodyera pubescens* R. Br. Downy rattlesnake plantain

(*Epipactis pubescens* (Willd.) A. A. Eaton; *Peramium pubescens* (Willd.) MacM.)

Most abundant in dry or moist coniferous woods, rather common in deciduous woods, especially so beneath rhododendron thickets in the mountains. June-August.

Mountain counties: Alleghany: *Correll* 4066 (D). Ashe: *Ashe* (NC); *Correll* 3962 (D). Avery: *Jony Cat.* 479 (U); *Correll* 3948 (D, MBD). Buncombe: *Beals* (HMD); *Standley and Bollman* 9984 (U); *Correll* 3838 (D, HMD), 215, 3809 (D). Burke: *Correll* 3898 (D, But.). Caldwell: *Small and Heller* (F). Haywood: *Eliason* (NC); *Oosting* 34573, 35298 (D); *Correll* 3809 (D). Henderson: *Blomquist* 5932 (D); *Oosting* 35483 (D); *Correll* 3254, 3315 (D), 3290 (D, GHG). McDowell: *Blomquist and Correll* 4675 (D). Macon: *Coker and Holland* (NC). Mitchell: *Wetherby* (HMD); *Correll* 3891 (D). Polk: *Peattie* 1068 (F, NC); *Day* (Da.); *Correll* 3212, 3254 (D). Rutherford: *Abbe and Spalteholz* (C); *Hulst* (B); *Blomquist and Correll* 5109 (D). Transylvania: *Correll* 3429, 3433 (D). Watauga: *Robinson* 100 (A); *Heller* (NS, N); *Huger* (N); *Randolph* 1156 (C, G); *Small and Heller* (C). Yancey: *Correll* 3879 (D).

Piedmont counties: Burke: *Correll* 3057 (D, POS). Catawba: *Correll* 2812 (D, HMD, A). Chatham: *Correll* 714 (D). Cleveland: *Correll* 3002 (D). Durham: *Creaser* (D); *Correll* 620, 2510, 3891, 4648 (D). Forsyth: *Schallert* (POS); *Schweinitz* (U); *Lehman* (S); *Correll* 2613, 4080 (D). Guilford: *Matthews and Ward* (NC); *Schallert* (D). Iredell: *Blomquist and Correll* 4671 (D). Montgomery: *Correll* 7040 (D). Orange: *Wallace* (NC); *Webb* (NC); *Totten* (NC); *Couch* (NC); *Norton* (U); *Blomquist* 5931 (D); *Millsaps* (NC); *Abbe and Spalteholz* (C); *Correll* 4721 (D). Person: *Oosting* 33341 (D). Polk: *Townsend* 3 (C). Randolph: *Correll* 664, 732 (D). Rutherford: *Correll* 3181 (D). Surry: *Correll et al.* 4948 (D). Warren: *Scholz* (NC).

Coastal Plain counties: Halifax: *Williamson* (NS).

Newfoundland to Florida, west to Alabama and Minnesota.

9. PONTIEVA R. BR. PONTIEU'S ORCHID

1. *Ponthieva racemosa* (Walt.) C. Mohr. Ponthieu's orchid

On the edge of woodland streams and ponds, or on the edge of muddy sloughs. September–October.

Coastal Plain counties: Brunswick: *Oosting* 33723 (D). Columbus: *Gibbes* (N). New Hanover: *Wherry* (N).

Virginia to Florida and Alabama; also in the West Indies and Central and South America.

10. MICROSTYLIS NUTT. ADDER'S MOUTH

(*Malaxis* Soland; *Achroanthes* Raf.)

Leaf 1, ovate, close-sheathing, spreading about midway up scape, 2–6 cm. long; lip green below, 3-lobed at apex, middle lobe smaller than the lateral pair, minute; flowers green 1. *M. unifolia*

Leaves 2, broad-ovate, sheathing scape at base, nearly opposite, 3–10 cm. long; lip brownish green above, 3-lobed at apex, lateral lobes shorter than the broad, blunt middle lobe; flowers green with orange or vermillion markings

2. *M. spicata*

1. *Microstylis unifolia* (Michx.) BSP. Green adder's mouth
(*Microstylis ophioglossoides* Nutt.; *Malaxis unifolia* (Michx.) BSP.;
Achroanthes unifolia (Michx.) Raf.)

In low moist acid soil and humus. June–August.

Mountain counties: Buncombe: *Forester* (N); *MacElwee* (NS); *Schallert* (D); *Williamson* (NS); *Robinson* 64 (G); *Hall* (B). Burke: *Heller* (P, NS); *Small and Heller* (U). Caldwell: *Small and Heller* (NS). Cherokee: *Correll* 3604 (D, GHG). Clay: *Oosting* 34590 (D). Henderson: *Correll* 3252, 3301A (D); *A. B. A.* (N). Jackson: *Correll* 3492 (D). Macon: *Smith* (U). Mitchell: *Wetherby* (HMD); *Correll* 3895 (D); *Ashe* 1327 (NC). Polk: *Peattie* 1076 (NC). Swain: *Wright et al.* 207 (C). Transylvania: *House* 3640 (G); *Oosting* 36115 (D); *Correll* 3413 (D). Watauga: *Heller* (N); *Ashe* (NC); *Robinson* (G); *Durand* (C); *Small and Heller* (F). Yancey: *Leeds* 1200 (NS); *Correll* 3878 (D).

Piedmont counties: Durham: *Billings* (D); *Correll* 4759, 4950 (D). Forsyth: *Schweinitz* (NS). Lee: *Totten* (NC); *Wright et al.* 208 (C); *Correll* 6969 (D). Moore: *Correll* 6998 (D).

Coastal Plain Counties: Beaufort: *Blomquist* 5951 (D). Bertie: *Lay* (NC). Bladen: *Fraser* (N). Johnston: *Deans* (NC); *Mitchell* (D). Onslow: *Ashe* (NC).

Newfoundland to Georgia, west to Manitoba, Louisiana, Wisconsin, and Illinois.

2. *Microstylis spicata* Lindl. Florida adder's mouth
(*Malaxis spicata* Sw.)

So far as the writer has been able to determine, no specimen exists and no report has been made concerning the occurrence of this orchid in North Carolina. Small gives the range from Florida to Virginia, where it occurs in the coastal plain. He says the species grows in "low or wet hammocks, and stream banks, often in calcareous soil" (20). May–July.

11. LIPARIS L. C. RICH. FALSE TWAYBLADE

Lip oblong-spatulate or obovate, 5 mm. long, about 4 mm. wide, abruptly pointed, yellowish green.....1. *L. Loeselii*
Lip broadly wedge-ovate, 10 mm. long or more, 7 mm. wide, mucronate, translucent, madder-purple.....2. *L. liliifolia*

1. *Liparis Loeselii* (L.) L. C. Rich. Loesel's Twayblade

Wet rocky soil and on cliffs in mountain woods, moist thickets and swampy soil. May–July.

Mountain counties: Rutherford: *Barksdale* (NC, D).

Maritime Provinces to District of Columbia, also mountains of North Carolina, west to Indiana, Wisconsin, and Minnesota.

2. *Liparis liliifolia* (L.) L. C. Rich. Lily-leaved twayblade

Low moist floodplain woods and in thickets along streams, or on rocky sandy soil on moist wooded slopes. May-July.

Mountain counties: Ashe: *Ashe* (NC). Avery: *Correll* 3942 (D). Buncombe: *Beals* (HMD); *Harshberger* (P); *Forster* (N); *Billmore* 566b (N); *Correll* 3847 (D). Burke: *Oosting* 365a (D); *Correll* 3915 (D). Caldwell: *Small and Heller* (F). Graham: *Correll* 3678 (D). Haywood: *House* 4491 (U); *Blomquist* 5949 (D). Henderson: *Campbell* (NC). Jackson: *Williamson* (NS); *Thaxier* (U, G). McDowell: *LeRoy and Ruger* (HMD). Macon: *Leeds* 1113 (NS); *Buckley* (N). Mitchell: *Huger* (N); *Wetherby* 147 (U); *Ashe* (NC). Swain: *Wright et al.* 205 (C). Transylvania: *House* 4272 (U).

Piedmont counties: Durham: *Correll* 547 (D, GHG). Forsyth: *Schweinitz* (NS); *Denke* (D). Granville: *Correll* 470 (D). Orange: *Couch* (NC); *Correll* 310 (D); *Mathews* (NC). Surry: *Rusby* (N).

Maine to Georgia and Alabama, west to Iowa, Missouri, Minnesota, Illinois, and Ohio.

12. CORALLORRHIZA (HALLER) R. BR. CORAL ROOT.

1. Lip 3-lobed.

2. Lip 3-4 mm. long, shallow lateral lobes below middle, middle lobe ovate, white, not spotted; spur obsolete 1. *C. trifida*
2. Lip 6-8 mm. long, distinctly hastately-lobed near base, middle lobe rounded at apex, suborbicular, purple blotches on face; spur rather prominent 2. *C. maculata*

1. Lip not 3-lobed, at most notched, eroded or wavy along margin

3. Lip 4 mm. long, 5 mm. wide, eroded on the margin, broadly rounded, abruptly contracted at the base, white, spotted with magenta-crimson 3. *C. odororhiza*
3. Lip 5-6 mm. long, 4-5 mm. wide, oval or suborbicular, notched at apex, margin entire or denticulate, white with purple spots 4. *C. wisteriana*

1. *Corallorrhiza trifida* Chatelain. Early coral root (*Corallorrhiza innata* R. Br.)

No specimen of this species from North Carolina has been seen, although the state is perhaps within its range. Memminger (16) included this species under the name *C. Corallorrhiza* (L.) Karst. in his list of Henderson County plants, and Curtis (13) catalogued it as *C. innata* R. Br. He gave its range as upland (mountain) districts. May-July.

Newfoundland and New England to North Carolina (?), west to Alaska, Washington, British Columbia, and Oregon.

2. *Corallorrhiza maculata* Raf. Spotted coral root (*Corallorrhiza multiflora* Nutt.)

In rich decaying humus in upland deciduous woods and along shaded stream banks, in slightly acid habitats. July–August.

Mountain counties: Ashe: *Correll* 4044 (D). Avery: *Robinson* (G); *Correll* 3950 (D). Buncombe: *Correll et al.* 3850 (D). Burke: *Correll* 3897 (D). Haywood: *Standley* 5392 (U). Mitchell: *Wetherby* (HMD). Swain: *Hulst* (B). Watauga: *Heller* (N). Yancey: *Correll* 3877 (D).

Piedmont counties: Forsyth: *Lehman* (S).

Newfoundland to Florida, west to Texas, California, and British Columbia.

3. *Corallorrhiza odontorhiza* (Willd.) Nutt. Autumn coral root

Moderately acid conditions in coniferous or deciduous woods, dry or moist soil, on slopes or in floodplain regions. August–October.

Mountain counties: Avery: *Robinson* (G). Buncombe: *Beals* (HMD); *Biltmore* 305 (U). Burke: *Blomquist and Correll* 4701 (D). Haywood: *Standley* 5587a (U). Henderson: *Campbell* (NC); *Shoolbred* (N). Jackson: *Huger* (N); *Williamson* (N, NS). McDowell: *Blomquist and Correll* 4676 (D, HMD). Madison: *Wherry and Pennell* 14260 (NS). Mitchell: *Wetherby* (HMD). Polk: *Peattie* 1489 (NC). Swain: *Beardslee and Kofoed* (N, C, G, F); *Hulst* (B). Watauga: *Durand* (C).

Piedmont counties: Durham: *Latham* (D); *Correll* 4739, 4745, 4949, 4650 (D), 4747 (D, HMD), 7450 (D, GHG), 4755 (D, POS). Forsyth: *Schallert* (S, POS); *Schweinitz* (NS). Mecklenburg: *Gray* (WV). Moore: *Correll* 6987 (D). Orange: *Totten* (NC); *Coker* (NC); *Ward* (NC); *Matthews* (NC); *Barksdale* (NC); *Oosting* 35629 (D); *Correll* 4731, 4904, 4741 (D), 4915 (D, A). Polk: *Townsend* (C). Wake: *Wells* (SC).

Coastal Plain counties: Beaufort: *Blomquist* 7956 (D). Richmond: *Correll* 7156 (D). Scotland: *Correll* 7171A (D).

Maine to Florida, west to Texas and Michigan.

4. *Corallorrhiza wisteriana* Conrad Wister's coral root

On damp rich wood floors and ravine slopes. April–May.

Coastal Plain counties: Halifax: *Williamson* (NS).

Pennsylvania to Florida, west to Texas, Missouri, and Indiana.

13. TIPULARIA LINDL. CRANE-FLY ORCHID

1. *Tipularia discolor* (Pursh.) Nutt.

(*Tipularia unifolia* (Muhl.) BSP.)

Rich damp woods and along shaded banks of streams, moderately acid soil. July–September.

Mountain counties: Alleghany: *Correll* 4073 (D). Buncombe: *Correll* 3837 (D, POS). Burke: *Heller* (F, P, N, NS); *Blomquist and Correll* 4700 (D). Cherokee: *Leeds* 1162 (NS); *Schweinitz* (NS). Graham: *Correll* 3669 (D). Haywood: *Holmes* (NC). Henderson: *Campbell* (NC); *Correll* 3316 (D); *Memminger* (NC).

Polk: *Day* (Da.); *Peattie* 987 (NC), 1030 (G); *Correll* 3190 (D). Rutherford: *Biltmore* 1041a (N); *Townsend* (U, C). Yancey: *Leeds* 1202 (NS).

Piedmont counties: Anson: *Correll* 7098 (D). Burke: *Correll* 3056, 3100 (D). Caldwell: *Randolph* 1083 (C). Catawba: *Correll* 2931 (D). Cleveland: *Correll* 3004 (D). Davie: *Blomquist and Correll* 4668 (D). Durham: *Blomquist* 5956, 7584 (D); *Oosting* 33238 (D); *W.B.D.* 25 (D); *Correll* 4652 (D, But.), 4749, 4730, 4960 (D). Forsyth: *Schallert* (POS, N); *Schweinitz* (NS); *Correll* 4095 (D, HMD, GHG, A). Franklin: *Oosting* 35623 (D). Gaston: *Correll* 2945 (D). Granville: *Correll* (D). Guilford: *Schallert* (D). Iredell: *Blomquist and Correll* 4669 (D). McDowell: *Blomquist and Correll* 4673 (D). Mecklenburg: *Abbe and Spalteholz* (C); *Correll* 3005 (D). Montgomery: *Correll* 7041 (D). Moore: *Correll* 7005 (D). Orange: *Totten* (NC); *Alexander* (NC); *Webb* (NC); *Millsaps* (NC); *Wallace* (NC); *Oosting* 33255 (D); *Correll* 4740 (D), 4908 (D, HMD, GHG). Rowan: *Correll* 2808 (D). Rutherford: *Correll* 3177 (D). Stanly: *Small and Heller* (N, F). Union: *Correll* 7075 (D). Warren: *Scholz* (NC).

Coastal Plain counties: Halifax: *Williamson* (NS, N); *Bartram* (NS). Johnston: *Deans* (NC). Washington: *Blount* (NC).

New Jersey and Delaware to Florida and the Gulf States, west to Ohio and Indiana.

14. EPIDENDRUM L. TREE ORCHIDS

(*Amphiglottis* Salisb.)

1. *Epidendrum conopseum* R. Br. Green-fly orchid, Florida epidendrum (*Amphiglottis conopsea* (Ait.) Small)

In swamps and marshy areas, mostly on sweet gum (12) and tupelo gum trees; farther south it has been found on live oaks, magnolias, and rarely upon rocks. July–September.

Coastal Plain counties: Columbus: *Rankin* (D, N); *Blomquist and Correll* 4900 (D, HMD, A, GHG, POS).

North Carolina to Florida, Alabama, and Louisiana.

15. CALOPOGON R. BR. GRASS-PINK

(*Limodorum* L.)

1. Flowers white, tinged with purple; middle lobe of lip 5–6 mm. wide, cuneate or wedge-shaped; column wings broadly triangular; leaf blades narrowly linear.....1. *C. pallidus*
1. Flowers bright purple, rarely white; middle lobe of lip larger than in the preceding species
2. Middle lobe of lip 9–10 mm. wide, broadly rounded at the base, orbicular; column wings nearly half-orbicular; leaf blades narrowly linear
2. *C. parviflorus*
2. Middle lobe of lip about 10 mm. wide, broadly fan-shaped or flabellate and truncate at apex; column wings rhombic; leaf blades linear, becoming slender lance-shaped.....3. *C. pulchellus*

1. *Calopogon pallidus* Chapm. Pale grass-pink*(Limodorum pallidum* (Chapm.) C. Mohr)

Often found growing with *C. pulchellus* in savannahs or wet open pinelands where intensely to moderately acid conditions exist. May-July.

Coastal Plain counties: Beaufort: *Brues* 5N (A); *Correll* 1586, 1587 (D). Bladen: *Blomquist and Correll* 2581 (D); *Correll* 1243 (D). Columbus: *Schallert* 241 (D, POS). Craven: *McCarthy* (U); *Correll* 1448 (D, HMD, GHG). Duplin: *Wherry and Trudell* (HMD); *Mathews* (NC); *Correll* 1321 (D, A, POS, But.). Johnston: *Deans* (NC). Moore: — (A). New Hanover: *Wright* (C). Onslow: *Moldenke* 6013 (A). Pender: *Wells* (NC, SC). Richmond: *Correll* 1113 (D). Sampson: *Correll* 1246 (D). Scotland: *Correll* 1174, 1172 (D).

North Carolina to Florida and Louisiana.

2. *Calopogon parviflorus* Lindl. Bearded calopogon*(Limodorum parviflorum* (Lindl.) Nash; *Calopogon barbatus* (Walt.) Ames)

In open savannahs, acid meadows, and low pinelands. April-May.

Coastal Plain counties: Brunswick: *Coker* (NC); *Totten* (NC); *Fogg* 5493 (P); *Blomquist* 5948 (D). Columbus: *Schallert* (POS, WV). Craven: *Leeds* 1648 (NS). Harnett: *Totten* (NC). New Hanover: *Bartram and Long* 1106 (NS); *Williamson* (NS); *Burke* (P); *Macfarlane* (P); *Canby* (A, F); *Churchill* (G). Onslow: *House* 5088 (N).

North Carolina to Florida, along the coastal plain.

3. *Calopogon pulchellus* (Sw.) R. Br. Grass-pink*(Limodorum tuberosum* L.)

In acid bogs and meadows, savannahs, and on the edge of open woods, strongly to moderately acid soils. May-July.

Mountain counties: Avery: *Ashe* (NC). Burke: *Small and Heller* (P, U, F, NS); *Heller* (N); *Gray et al.* (NS). Graham: *Oosting* 34633 (D). Haywood: *Oosting* 34729 (D). Henderson: *Wherry* (HMD); *Shoolbred* (N); *Biltmore* 313d (N); *Campbell* (NC); *Blomquist* 5925 (D); *Correll* 3330 (D). Jackson: *Huger* (N); *Williamson* (NS); *Oosting* 35343 (D, MBD). McDowell: *House* 4308 (U). Macon: *Wilson and Underwood* 2694 (T); *Wilson, Underwood, and Lott* 2782 (T); *Harbison* 16 (G, HMD). Mitchell: *Ashe* (NC). Transylvania: *House* 4379 (U, F, G). Watauga: *Ashe* (NC).

Piedmont counties: Anson: *Correll* 1036 (D). Catawba: *Small and Heller* (F, N); *Heller* (N, G). Forsyth: *Schweinitz* (NS); *Schallert* (S). Gaston: *Coker* (NC). Richmond: *Correll* 1105 (D). Stokes: *Ashe* 17 (NC). Wake: *Coit* 1250 (SC).

Coastal Plain counties: Beaufort: *Brues* (A); *Correll* 1688 (D, POS), 1586A (D). Bladen: *Blomquist and Correll* 2580 (D); *Correll* 401 (D). Brunswick:

Leeds 1135 (NS). Columbus: *Schallert* (POS, D); *Heller* (F). Craven: *Brown* 27 (U); *Shrink* (U); *Correll* 1462 (D). Cumberland: *Totten and Harbison* (NC). Dare: *Correll* 1725 (D). Duplin: *Wherry and Trudell* (HMD); *Chapman* (NS); *Mathews* (NC); *Correll* 1586A (D). Hoke: *Correll* 1115 (D). Hyde: *Correll* 1759, 1774 (D). Johnston: *Deans* (NC). Moore: *Meritte* (L.); *Katzenstein* (G); *Wright et al.* 243 (C). New Hanover: *Biltmore* 313a (N); *Ward* (N); *Bartram* (NS); *Wright* (C). Pamlico: *Correll* 1486A (D, A, But.). Pender: *Wells* (NC). Richmond: *Correll* 1105 (D). Robeson: *Correll* 1234 (D). Sampson: *Correll* 1301 (D). Scotland: *Correll* 1173 (D, GHG, HMD, A). Washington: *Wiegand and Manning* 891 (C). Wilson: *Blomquist* 7711 (D).

Newfoundland and Nova Scotia to Florida, Alabama, Louisiana, and Texas, westward to Minnesota and Missouri.

3a. *Calopogon pulchellus* f. *albiflorus* (Britton) Fernald White grass-pink

The flower in this form is pure white with cream-colored hairs on the face of the lip. The plant is smaller than the species. In open savannahs and moist sphagnum bogs. May-July.

Coastal Plain Counties: Brunswick: *Leeds* 1133 (NS). Duplin: *Correll* 1318 (D). New Hanover: *Wright* (C). Robeson: *Correll* 1234A (D).

Distribution similar to that of the species.

16. HEXALECTRIS RAF. CRESTED CORAL ROOT

1. *Hexalectris spicata* (Walt.) Barnhart Crested coral root
(*Hexalectris aphylla* (Nutt.) Raf.)

In open rocky woods and rich woods along streams, in moderately acid habitats. July-August.

Mountain counties: Swain: *Beardslee and Kofoid* (P, A, N, G, WV, F).

Piedmont counties: Davidson: *Totten* (NC). Durham: *Correll* 4651 (D). Forsyth: *Schweinitz* (NS); *Schallert* (POS); *Correll* 4078 (D). Orange: *Alexander* (NC); *Blair and Totten* (NC). Rowan: *Small and Heller* (F). Stanly: *Small and Heller* (N).

Coastal Plain counties: Johnston: *Deans* (NC). New Hanover: *Wood* (U).

Maryland to Florida and the Gulf States, west to Texas, Arizona, Missouri, and Indiana.

• 17. APLECTRUM (NUTT.) TORR. PUTTY ROOT

1. *Aplectrum hyemale* (Muhl.) Torr. Putty root
(*Aplectrum spicatum* (Walt.) BSP.)

In mucky, wet soil in wooded floodplains, or in rich low moist woods, neutral to slightly acid soils. May-July.

Mountain counties: Buncombe: *Biltmore* 1287 (N). Polk: *Schiele* (NC). Yancey: *Biltmore* 1287 (N).

Piedmont counties: Catawba: *Correll* 2930 (D). Durham: *Correll* 5013 (D). Forsyth: *Schallert* (POS); *Schweinits* (NS); *Denke* (S); *Lehman* (D); *Correll and Butts* 5026 (D); *Correll* 4096 (D). Guilford: *Schallert* (D). Orange: *Totten* (NC); *Womack* (NC); *Blomquist* 5922 (D); *Correll* 5010 (D).

Ontario and Vermont to Georgia, west to California and Saskatchewan.

18. EULOPHIA R. BR.

(*Triorchos* Small and Nash)

1. *Eulophia ecristata* (Fernald) Ames

(*Triorchos ecristatus* (Fernald) Small)

Pinelands and scrubs, open rocky soil, rather dry. August.

Coastal Plain counties: New Hanover: *Williamson* (NS).

Other collections: "Mountains of North Carolina, August 1892": *Williamson* (F). "North Carolina, 1889": *McCarthy* (N).

Florida and North Carolina.

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LITERATURE CITED

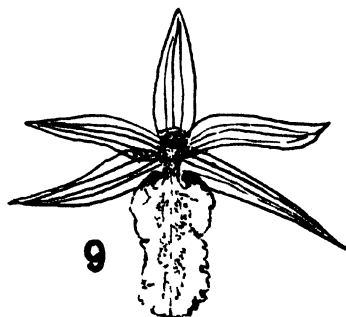
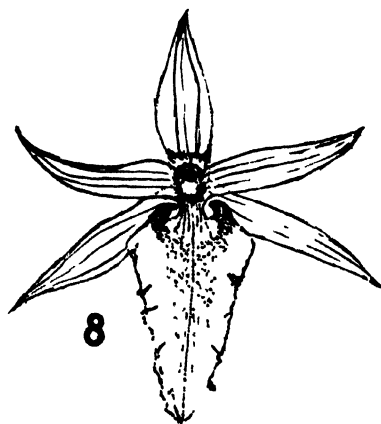
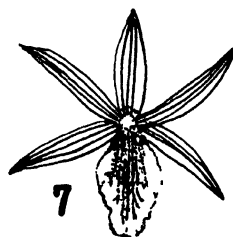
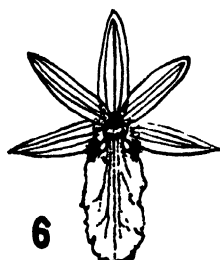
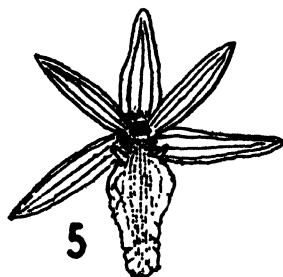
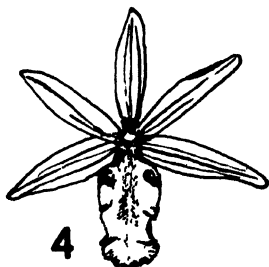
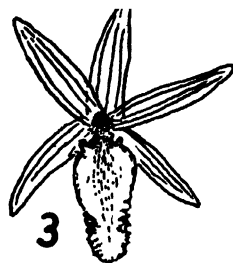
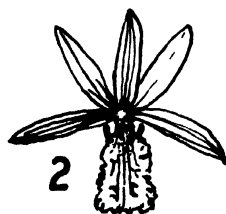
1. AMES, OAKES. 1903. Natural hybrids in *Spiranthes* and *Habenaria*. *Rhodora* 5: 261-264.
2. ———. 1905. Contributions toward a monograph of the American species of *Spiranthes*. *Orchidaceae*, Fascicle I: 113-156.
3. ———. 1910. The genus *Habenaria* in North America. *Orchidaceae*, Fascicle IV: The Merrymount Press. Boston.
4. ———. 1921. Notes on New England orchids—*Spiranthes*. *Rhodora* 23: 73-85.
5. ———. 1924. An enumeration of the orchids of the United States and Canada. The American Orchid Society. Boston.
6. ANDREWS, A. LEROY. 1901. A natural hybrid between *Habenaria lacera* and *H. psychodes*. *Rhodora* 3: 245-248.
7. BARKSDALE, LANE. 1933. Orchid hunting in Guilford County. *The High School Journal* 16: 232-241.
8. ———. 1936. Some notes on orchids of the Piedmont and western North Carolina. M. A. Abernethy, Publisher. Chapel Hill.
9. BLOMQUIST, H. L., AND H. J. OOSTING. 1936. A guide to the spring and early summer flora of the Piedmont, North Carolina, pp. 43-46. Seeman Printery, Durham.
10. BRITTON, N. L., AND A. BROWN. 1896. An illustrated flora of the Northern United States, Canada, and the British Possessions. Ed. 2, Vol. 1: 456-481. Charles Scribner's Sons. New York.
11. CHAPMAN, A. W. 1897. *Flora of the Southern United States*. Ed. 3. American Book Co. New York, Chicago, Cincinnati.

12. CORRELL, DONOVAN S. 1936. *Epidendrum conopseum* in North Carolina. Jour. Elisha Mitchell Sci. Soc. 52: 91-92.
13. CURTIS, M. A. 1867. Botany: containing a catalogue of the indigenous and naturalised plants of the state, pp. 53-54. Geol. and Nat. Hist. Survey of North Carolina. Part 3.
14. EAMES, EDWARD A. 1921. An unusual form of *Habenaria clavellata*. Rhodora 23: 126-127.
15. HYAMS, M. E. 1884. A preliminary list of additions to Curtis' catalogue of indigenous and naturalised plants of North Carolina—flowering plants. Jour. of the Elisha Mitchell Sci. Soc. 2: 72-76.
16. MEMMINGER, E. R. 1915. List of plants growing spontaneously in Henderson County, North Carolina. Jour. of the Elisha Mitchell Sci. Soc. 30: 126-149.
17. PFITZER, E. 1889. *Orchidaceae*. From A. Engler and K. Prantl *Die Naturalischen Pflanzenfamilien* II: 52-218.
18. ROBINSON, B. L., AND M. L. FERNALD. 1908. Gray's New Manual of Botany, pp. 304-319. American Book Co. New York, Chicago, Cincinnati.
19. SCHWEINITZ, LEWIS DAVID VON. 1821. "Flora Salemitana." Unpublished manuscript in possession of the Academy of Natural Sciences. Philadelphia.
20. SMALL, J. K. 1933. Manual of the Southeastern flora, pp. 363-399. Small. New York.
21. SMALL, J. K., AND A. A. HELLER. 1892-95. Flora of the western North Carolina and contiguous territory. Torrey Bot. Club Mem. 3: 1-39.
22. WIEGAND, K. M. 1899. A revision of the genus *Listera*. Torrey Bot. Club Bull. 26: 157-171.
23. WOOD, THOMAS F., AND GERALD MCCARTHY. 1883-86. Wilmington flora, New Hanover County. Jour. of the Elisha Mitchell Sci. Soc. 3: 77-134.

EXPLANATION OF PLATE 15

- Fig. 1. Lip of *Spiranthes ovalis*. × 4.
- Fig. 2. Lip, petals, and sepals of *Spiranthes Beckii*. × 4.
- Fig. 3. Lip, petals, and sepals of *Spiranthes floridanum*. × 4.
- Fig. 4. Lip, petals, and sepals of *Spiranthes gracilis*. × 4.
- Fig. 5. Lip, petals, and sepals of *Spiranthes longilabris*. × 2.
- Fig. 6. Lip, petals, and sepals of *Spiranthes praecox*. × 2.
- Fig. 7. Lip, petals, and sepals of *Spiranthes vernalis*. × 2.
- Fig. 8. Lip, petals, and sepals of *Spiranthes odorata*. × 3.
- Fig. 9. Lip, petals, and sepals of *Spiranthes cernua*. × 2.

PLATE 15



STUDIES IN THE LIFE HISTORY OF NITELLA HYALINA AGARDH

By LAURIE M. STEWART

PLATES 16-19 AND ONE TEXT FIGURE

INTRODUCTION

Due to their unique structure and reproductive cycle, the Charales have for many years aroused the interest of botanists and other men of science, and for the same reasons have caused much disagreement as to their probable phylogenetic position. Accordingly, the group has at one time or other, been placed in every phylum of the plant kingdom (25). The generally accepted idea at present is that they are a very highly specialized off-shoot of the Chlorophyceae.

Adequate reviews of the work done on the classification of the group are presented by Allen (2) and Robinson (25). Karling (17) has quite thoroughly summarized the important cytological contributions made toward a better understanding of the Charales. References to practically all the important papers that have been written on any phase of the subject are to be found compiled by Fritsch (10) in his literature of the Charales.

It is apparent that most of the work has been done on the single genus *Chara*, while the genus *Nitella* has been somewhat neglected (17). The purpose of the present paper is to study as completely as possible the life cycle of a single species of *Nitella*, in order (1) to check the development of the reproductive organs against that described by previous investigators for the Charales, (2) to secure a chromosome count for the species, and (3) to determine if possible, where reduction division takes place in this species.

DESCRIPTION OF THE PLANT

The following description of the plant is based wholly on material grown in an aquarium as described under "Materials and Methods" of the present paper. The determination of the species as *N. hyalina* was made by following the key in *The British Charophyta*, Vol. I, by Groves and Bullock-Webster (14). Up to the present time the writer

has found no record of the collection of this species in North America, although Groves and Bullock-Webster state (without reference to author or locality) that it has been found in this country. *Nitella hyalina* is not listed in Allen's *Characeae of America*.

Nitella hyalina Agardh

Plants homothallic, 10 to 27 or even 30 cm. long, ascending from clumps in a somewhat upright, more or less flexible or curving manner (Text fig. 1). Stems 250–290 μ in thickness; lower internodes elongated, upper ones increasingly shorter, usually with denser verticils. A very distinctive feature is the presence of a smaller whorl or verticil within the larger primary verticil, the former usually containing twice the number of laterals of limited growth as the latter (Plate 16).

Primary verticils consist most often of 8 laterals which are typically 3-times divided, though 4th divisions are sometimes found. The basal nodes of the laterals are cut off close to the nodal cells of the main axis or stem, and from these arise the laterals of the smaller whorl. The second node of each lateral produces 6 (5–8) leaflet rays which frequently cut off a node and form 5–6 rays, each terminated by a mucro. Sometimes several of these leaf rays produce still another node and a whorl of from 3–5 rays. The mucros are usually 32–43 μ broad by 72–74 μ long (Plate 19, fig. 2).

Whorls of secondary laterals (Plate 16) numbering from 8–16, usually 16, are frequently found at each stem node. These laterals are usually from $\frac{1}{3}$ to $\frac{2}{3}$ the length of the primary laterals and consist of either an undivided or a once-divided leaf, always terminated by a mucro. The divided laterals bear organs of both sexes.

Antheridia and oogonia are borne at the nodes, and all nodes except the basal ones have been found fertile. Antheridia 200–300 μ in diameter; pale yellow-green when young, changing through yellow-orange to red-orange when mature. Terminating all divisions of the leaf except the ultimate rays. Always borne singly.

Oogonia single (rarely 2) at every node except the basal one. Entire oogonium 516–590 μ long by 338–410 μ thick. Mature oospore 249–290 μ ; wall deep reddish-brown when mature; surface minutely pitted; ridges prominent, 7–8.

The University Lake species differs from the *Nitella hyalina* as described by Groves and Bullock-Webster chiefly in having shorter mucros and smaller antheridia. These slight variations do not seem sufficient to warrant the giving of any form name to the Lake species.

A study of Allen's key (2) reveals the fact that similar variations frequently occur in many species of *Nitella*.

MATERIALS AND METHODS

Living plants for the present study were collected from University Lake, Chapel Hill, N. C., and planted in a south window aquarium in which a previous culture of the same plant had been growing since January 1936. The cultures were in direct sunlight till about noon each day, and the room was usually warm both day and night.

At the time of collection the new material was not in fruit, and the plants did not assume a healthy appearance until November. During the cloudy days of December 1936 and January 1937, artificial light was used almost constantly for about three weeks. By the middle of January the plants were observed to be fruiting sparingly, and a month later nearly all the verticils were thick with fruit.

Material of all stages was fixed in Flemming's weak and Nawaschin's fluids and followed by Heidenhain's haematoxylin or Gram's modified stain. Sectioned material did not prove to be very satisfactory. Whole material stained by the Feulgen technic and teased out on the slide before mounting gave excellent results and this method was used almost exclusively for permanent mounts. The improved Feulgen technic as given by Tomasi (28) was followed with only slight modifications. The fixative used was prepared as follows: to 50 cc. of 6% aqueous HgCl_2 was added 1 cc. of glacial acetic acid, and the mixture heated to boiling, removed from the flame, and when bubbling ceased, was poured over the fresh material. After treatment in the fixative for 15 minutes, the material was washed for an hour in running water, then hydrolyzed for 4 minutes in a normal solution of HCl at 58°C . and finally stained for 3 hours.

Temporary mounts of antheridial filaments were made with iron-aceto-carmine as suggested by Karling (17) and studied for stages in antheridial development. Living material was also studied after staining with congo red, which rendered the walls, cytoplasm and nuclei quite visible without causing death before 7-9 hours. Methyl-violet and fuchsin killed, but very clearly and quickly stained the walls, cytoplasm and especially the nuclei. This stain was also used for studying the flagella of the sperm while in the mother cells and after escape.

VEGETATIVE STRUCTURE

The cell wall of the plant is quite rigid and not easily punctured because of the slippery surface which may be due to the presence of a

gelatinous substance (10). The wall stains well with methyl-violet and fuchsin. While apparently smooth on the outside, the wall has irregular thickenings on the inner surface, which show up plainly in sectioned material stained by the Feulgen technic. The single layer of oval chloroplasts are found in practically every cell of the plant. They are embedded in the peripheral layer of the parietal cytoplasm, and show clearly through the hyaline cell wall (Plate 19, fig. 2). In the elongated internodal cells they are arranged in fairly regular rows more or less parallel with the long axis of the stem; in the tip cells the scattered chloroplasts are pale green for a short time, but soon become colorless. The plastids of the manubria (Plate 17, fig. 15) are also arranged in broken lines and are pale yellow.

Just beneath the peripheral cytoplasm is the stream of rotating cytoplasm (Plate 19, fig. 3). Small and large granules move along in the stream, with much larger masses which are the nuclei. Some of these masses are partially constricted in the middle, due to the fact that the nuclei are in the process of amitotic division (Plate 19, fig. 12).

Streaming of the cytoplasm in the plant under observation has been seen in the following cells: all nodal cells, internodal cells, when fairly mature, basal and stalk cells of oogonia and antheridia, spiral sheath cells of the oogonia, rhizoids, and tip cells of the leaflets.* The smaller particles move more rapidly than the nuclei, which slow up noticeably when making the turn at either end of the cell. In the nodal cells of the stem, the chloroplasts, which are rounder and larger than those of the internodes, move around in the cytoplasm of the cell. In other parts, the chloroplasts are stationary. A verticil cut from a plant December 2, 1936, and kept in a petri dish of water by a north window, still shows actively flowing cytoplasm at the present writing, a period of about six months.

VEGETATIVE REPRODUCTION

Many of the Characeae reproduce by vegetative means (10). New axes of growth arise from secondary protonema, from the nodes of buried stems or broken verticils, and from tuber-like growths and bulbils (21). Pieces of old plants found detached in the grass and mud a few inches under water in University Lake, April 3, 1937, showed new stems growing from old nodes, and rhizoids coming from the same nodes with the stems. Rhizoids seem to be capable of appearing

* The laterals of limited growth are commonly called "leaves," and their ultimate divisions, "leaflets."

at any node that is under mud. One instance was observed in which starch had accumulated in quantity at a verticil node, and from these cells rhizoids had grown out. When left in water and open to the light for about two weeks, the first sections of the rhizoids showed typical stationary chloroplasts, as in the internodes.

DEVELOPMENT OF THE ANTHERIDIUM

Antheridia have been found at every node of the leaf except the basal node. A study of very young verticils reveals the fact that the antheridial initial stages occur quite early in the development of the laterals. The antheridia are terminal on the internodal cells of the leaves. The apical cell of an antheridium cuts off one basal cell (Plate 17, fig. 1), then assumes a spherical shape. The first two divisions of this apical cell are vertical (Plate 17, fig. 3) and at right angles to each other, making four cells. The third division is in a horizontal plane, and results in the eight-celled stage (Plate 17, fig. 4). Now, each of the eight cells divides by two periclinal walls (Plate 17, fig. 5), into a series of three cells, forming a total of twenty-four cells from the original apical cell. About this time, a second basal cell is cut off at the base of the first one. The twenty-four cells are arranged in the form of three concentric spheres. The eight cells of the outer sphere do not divide again, but form the shell of shield cells within which are other sixteen cells. The shield cells have interlocked edges (Plate 17, fig. 7). The eight cells of the inner sphere elongate radially and without further division become the eight cylindrical manubria (Plate 17, fig. 6). Each manubrium is attached distally to a shield cell and proximally to one of the eight innermost cells, the capitula. Each capitulum divides one or more times to form secondary and even tertiary capitula (18), and from these cells arise the initial cells of the spermatogenous filaments. By repeated division of cells throughout their length, the filaments become greatly elongated until they form a tangled mass of threads that completely fills the hollow sphere bounded by the shield cells. In the early stages the shield cells are sparsely filled with granules that are at first colorless, but which soon become pale yellow. As the antheridium approaches maturity, the granules turn red, so that the ripe male organ is a bright red-orange ball. The manubria also contain a few granules; the capitula sometimes have a few, but the filament cells are always free from chromoplasts. Rotation of cytoplasm has been observed only in the stalk and basal cells of the antheridium.

The development of the sperm has been rather thoroughly investi-

gated by Belajeff (10), Lindenbein (20), Mottier (22), Walther (31), and others, with resulting differences of opinion on certain points, notably, the origin of the flagella. Briefly, from my own observations, the development of the sperm appears to be as follows: the very young cells of the filaments are somewhat rounded and plump, and filled nearly to the walls by their single nuclei (Plate 17, fig. 8). Successive divisions of these cells (Plate 17, fig. 9) cause increase in the length of the filaments and the resulting cells are narrower and the nuclei are much less spherical than in the original cells (Plate 17, fig. 10). The final divisions give rise to thin discoid cells (Plate 17, fig. 11), the sperm mother cells proper. Each sperm mother cell has a single nucleus and gives rise to a single bi-flagellate sperm. The writer has not made a special study of the origin of the flagella. Stages in the *Nitella hyalina* under observation have been found repeatedly showing the mother cell nucleus shifting to one side of the cell, becoming very flat and discoid, then thinning in the middle and assuming an incomplete band formation (Plate 17, fig. 11). The further lengthening of the nucleus results in the formation of a long spiral with a pointed anterior end and a blunt posterior end. The two flagella seem to be attached a short distance from the anterior end. By staining with methyl-violet and fuchsin, the flagella can be clearly seen when the nuclei are becoming band-shaped, and have the appearance of fine wire coiled about the sperm body (Plate 17, fig. 12).

The method in which the antherozoids of the Characeae escape from the cells of the filaments seems to be in dispute. Fritsch (10) states that the sperm are liberated "by the falling apart of the shields of the antheridial wall and the subsequent gelatinisation of the walls of the mother-cells." It is not clear as to whether he means the softening of a part of the wall in certain restricted areas or the disintegration of the entire cellular structure. Roze (26) gives a different view in stating that "the mother cells are perforated for the passage of the bodies, the cell walls persisting, in contradiction to the other Cryptogams where the release of the antherozoids is effected by the dissolution of the cell wall." Groves and Bullock-Webster (14) quote M. Leon Guignard as saying that the sperm, when fully mature, burst from their cells and swim about in the water. Although the escape of living sperm has not been observed during the present study, two instances have been found in which the entire sperm body appeared to be still within the cell wall (Plate 17, fig. 13), while the flagella were free from the wall and whipping about vigorously in the surrounding water. In

several other instances, strands of empty cells have been observed with now and then a motionless sperm apparently held by its posterior end in the cell, but otherwise free and with the flagella in normal position. After repeated observations, the writer is inclined to favor the explanation of Roze, that the cells are perforated for the escape of the sperm. Such a conclusion seems justifiable by the facts that the empty cells maintain their original shape, size, and continuity of wall long after the sperm have escaped, and that staining of the empty cell walls reveals no obvious breaks. The same observations also make it impossible for the writer to accept the gelatinisation theory as here interpreted.

The free sperm are very long (Plate 17, fig. 14), and have quite long flagella. The sperm body measures 72μ .

DEVELOPMENT OF THE OOGONIUM

Various names have been applied to the female organ and its component cells. Allen (2) introduces the term *sporophydium* to include all the cells of the female organ, namely, the basal, nodal, sheath, and egg cells. He uses the term *oospore* to mean the fertilized egg with its dense cytoplasm full of starch and oil. The sheath cells he calls the *sporostegium* or *spore-capsule*. The spore is the cell which gives rise to the egg and the one to three little cells at its base, Braun's famous "Wendungszellen." Fritsch uses the term *oogonium* to signify the uppermost cell in the row of three that forms the initial stages of the female organ. This cell later cuts off one cell in *Chara* and three cells in *Nitella*, the aforementioned "Wendungszellen." Thus, Fritsch's term includes the egg and the cut-off cells. He uses no term to include all the cells that arise from the initial cell of the female organ. The word *oogonium* is here used to mean the same thing that is meant by Allen's *sporophydium*. The cell called the *oogonium* by Fritsch and others is the *primary oocyte* of Tuttle (30), who calls the entire organ the *oogonium*. In the present study, the terms *primary oocyte* and *oogonium* will be used in the manner that Tuttle has adopted.

Although the development of the oogonium in *Nitella hyalina* from University Lake is largely in accordance with that given by Fritsch and others, for other species of *Nitella* and *Chara*, several deviations do occur, which if not of great significance, are non-the-less interesting. A brief account of this development follows.

The oogonial initial is homologous with the apical cell of any leaflet in the whorl. By transverse division of the initial cell, a row of four

cells is cut off (Plate 18, fig. 1). The apical cell is the primary oocyte, supported by the nodal cell, beneath which are the two basal cells. The node soon cuts off five peripheral cells, the sheath primordia, so that a disk of six cells is formed between the oocyte and the basal cells (Plate 18, fig. 4). Sachs (27) and Fritsch figure only one basal cell, but in the species studied, the writer has consistently found two basal cells. Even before the origin of any sheath primordia, the oocyte begins to cut off the first of the three cells (Plate 18, fig. 2). The first cell is stated by Fritsch to be cut off by a wall that is more or less vertical to the base of the oocyte, but in every case observed in the species studied, the wall is curved at a definite 45° angle with the long axis of the row of primary cells (Plate 18, fig. 2). It is always cut off on the adaxial side. This cell corresponds to the *first polocyte* of Tuttle (29), but since the chromosome counts in the vegetative and reproductive cells in *Nitella hyalina* do not bear out Tuttle's contention that reduction of chromosome number occurs during the first division of the oocyte, the term *polocyte* cannot be correctly applied to the cell in this species. A second cell is soon cut off from the oocyte, oftentimes before the sheath primordia have become at all elongated (Plate 18, fig. 3). The wall of the second cell slopes obliquely away from the middle wall of the first cell, and is not vertical as stated by Fritsch and others (Plate 18, fig. 4). The third cell is cut off from the oocyte sometime later than the first two, after the sheath cells have become about twice the length of the oocyte, and the coronal cells have been formed. The wall of the third cell is definitely horizontal, and the cell lies at the base of the oosphere, and touches the wall of the second little cell (Plate 18, fig. 6).

After the three cells are cut off, the oosphere, which has already become quite large, elongates rapidly and increases in size, pushing past the three little cells, and causing them to bulge out like blisters toward its lower side (Plate 18, fig. 7). Although these three cells appear to have definite walls separating them from each other and from the oosphere, they are still within the original wall of the primary oocyte. Tuttle (29) states that in his plant the three cells are not cut off by successive divisions of the primary oocyte, but that the first cell cut off migrates toward the base and on its way divides to form two polar bodies, during the time the oocyte nucleus is cutting off a second cell. Groves and Bullock-Webster (14) state on pages 52-53, vol. I of their *British Charophyta*, that "at the base of the oogonium of the *Nitellae* are found three small cells called by Braun 'turning cells' (Wendungs-

zellen). The formation of the first of these cells originates at the apex whence it makes its way downwards (as Goetz shows in a series of illustrations) and finally divides into three cells at the base of the oosphere." These authors have apparently misquoted Goetz, who does not say that the first cell divides to form three cells, but rather plainly indicates that they are formed by the successive divisions of the large central cell (13, p. 3). In the present study, mitotic figures have been found which tend to show that these divisions are successive, and not as described by Tuttle or Groves and Bullock-Webster. Braun's term "Wendungszellen" cannot be applied to these cells in the present study if it be interpreted as meaning "wandering cells" or even "turning cells," for the following reason: the first of the three cells is cut off at the apex of the oocyte, and assumes a lateral and finally a basal position simply due to the fact that it is pushed aside by the growing oosphere. All three cells are really fixed in position throughout their history, and the movement toward the base is only relative. Similar observations were made in 1929 on *Nitella batrachosperma* by Walthers (31). The above conclusions, however, were arrived at before the writer had seen Walthers' paper.

After the three cells are cut off, the oosphere elongates until it becomes finger-shaped, and the sheath cells spiral tightly around it so that the young oogonium is for a short time of nearly uniform diameter throughout its length. Later, the oosphere becomes more or less spherical and the spirals more nearly horizontal. The coronal cells are at this stage set close to the oosphere. In connection with the origin of the coronal cells, a mitotic figure was found which shows the lower tier being cut off from the basal cell (Plate 18, fig. 12), which method is also true for *Nitella batrachosperma* as shown by Walthers (31). Bullock-Webster and Groves state that in the *Nitellae*, the lower tier is cut off from the upper one.

When the oosphere is ready for fertilization, the upper ends of the sheath cells elongate and form a chamber above the apex of the oosphere. Swelling of the elongating portions a short distance below the crown, divides the chamber into two hour-glass compartments, the upper being very small and connected with the larger lower one by a narrow channel. The slight separating of the five sheath cells just beneath the crown forms five slits for the entrance of the sperm (Plate 18, fig. 8). The receptive spot is at the apex of the oosphere. Anthidia with opening shields are usually found at the same nodes with oogonia ready for fertilization, and nearly all the organs of both sexes on nodes of the same age mature at the same time.

GERMINATION

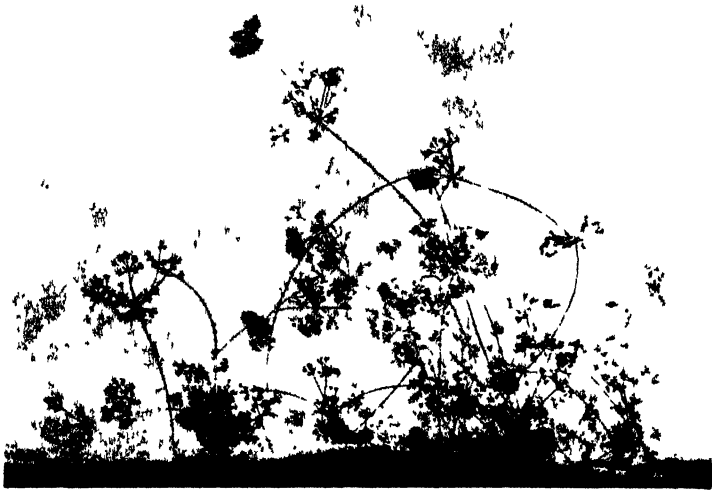
The process of germination has been very carefully worked out by Nordstedt, whose investigations were later reviewed and enlarged upon by the studies of de Bary (2). Although attempts to germinate the oospheres of the plant from the Lake have been so far unsuccessful, the oospheres have germinated in the aquarium. On January 9, 1936, a few plants from an old aquarium were put into a new aquarium. The plants were fruiting at the time of transplanting. About the middle of February, 1936, an irregular ring of young plants was noticed around the old clump. One of these was carefully taken up and found to have the old spore case still clinging to the base of the shoots, and in the upper verticils were found nearly mature antheridia and oogonia. The young plants were from 6-8 inches high. Other instances of germination have been found during the present spring. Plants from a previous collection were discarded from an aquarium, and the sand scraped off and replaced by fresh; new material from the Lake was transplanted October 17, 1936. This new material was not fruiting at the time of collection. The plants were again removed December 11, 1936, to another aquarium with an entire change of sand and water in which no plants had been growing. Fruit began to appear about the middle of January, 1937, and about a month later was abundant. About the first of March, mature antheridia were found with open shields, and on the same nodes were oogonia with elongated sheath cells. Black oospores were first seen about the 6th of April, and young plants 20 mm. in length were pulled up April 19th and found to be growing from oospore cases (Plate 19, fig. 4). It seems that it may be safely assumed that the young plants came up from oospores maturing since the middle of January, since it is not very likely that oospores maturing in the summer of 1936 would still be found in the scant soil around the rhizoids of the Lake material, especially after two separate transplantings. The protonemal threads of these new plants were found to be two-jointed, and lined with chloroplasts as are the internodes (Plate 19, fig. 4).

STUDY OF THE NUCLEI

The nuclei of *Nitella hyalina* present a wide range of sizes, shapes and positions that is quite interesting. A brief discussion of the several types follows:

Nuclei of internodal and leaf cells. The nuclei of very young cells are usually spherical and centrally placed. The cells are highly vacuo-

late and no sign of rotating cytoplasm is seen in the early stages. Later the vacuoles unite to form one large central vacuole around which the free cytoplasm rotates, carrying with it the one or more nuclei. The leaf and internodal cells are at first all uninucleate (Plate 19, fig. 1), but later become multinucleate by fragmentation or amitosis. The resulting nuclei are very small and more or less oval in shape, and not densely granular (Plate 19, fig. 2). All the nuclei move close to the walls (Plate 19, figs. 18 and 2).



TEXT-FIG. 1

Nuclei of tip and nodal cells. Each tip cell (muero) has only one nucleus (Plate 19, fig. 1), which is 7μ in diameter in fixed material, spherical and densely granular. It takes a deep stain when treated with Feulgen. The slow movement of the cytoplasm in the cell does not appreciably affect its position; it is usually basal.

The nodal cells are also uninucleate (Plate 19, fig. 10). The nucleus is centrally placed and is spherical and stains deeply with Feulgen, as do the tip cell nuclei.

Nuclei of the oogonia. A nearly mature oogonium stained by the Feulgen technic and mounted whole presents a very striking figure. The cells are faintly outlined in yellow, but only the nuclei take the

bright red stain. Near the base are the three large round nuclei in a row, the two basal and one nodal nuclei. Three smaller dots in a crescent at the base of the egg belong to the three little cells cut off by the oocyte. At the apical end of the oogonium are ten dark red dots, the coronal cell nuclei, and spiralling around the oospore are five long ribbons; these five threads have been called nuclei by the writer, after numerous observations on their development in both living and stained material. No references nor illustrations have been found in the literature concerning the nature of the nuclei in the sheath cells, with the exception of one figure 13b by Walthers (31) which shows a decidedly oval nucleus in a sheath cell which is just beginning to spiral. Bullock-Webster and Groves state that the cytoplasm in the sheath cells of mature oogonia maintain the form of coils and can be unwound and laid out in separate strands with the integument. After many observations, the writer is still inclined to believe that the coils which show up so clearly with Feulgen in *Nitella hyalina* are true nuclei, and not cytoplasmic strands. It does not seem likely that the cytoplasm in the sheath cells would stain at the same time that the cytoplasm in every other cell remains colorless. Cross-sections of nearly mature oogonia that have been previously stained with Feulgen, show the sheath cells as rings tangent to the circle of the egg. In many of these rings is found a round, granular, Feulgen-stained body of the same diameter and structure as the ribbons in the spiral cells previously mentioned. This body is usually embedded in a granular substance which is light yellow in color and gives the same appearance as cytoplasm in other cells of the plant (Plate 18, fig. 8a). Also, under other staining processes, the cytoplasm is not found in bands in the cells of the sheath. The development of these sheath cell nuclei is as follows: each primordium has a single, spherical nucleus which is central in position (Plate 18, fig. 9). After the primordium has cut off two coronal cells, the lower of the three cells in the thread elongates and begins to spiral, and its nucleus also elongates and spirals with it (Plate 18, fig. 10). As this process of elongation continues, the sheath cell becomes very long, and the nucleus finally extends from the base of the cell to within $\frac{1}{3}$ the distance from the tip of the coronal cells (Plate 18, fig. 11). Nuclei in the sheaths of mature oogonia measure 7μ by 600μ . Incidentally, these measurements also represent the range in length of the nuclei found throughout the plant body, from the 7μ diameter tip cell nuclei to the 600μ sheath cell nuclei.

CHROMOSOME COUNT

Chromosome counts were attempted in the following cells: sheath primordia, coronal cells, antheridial filament cells, the 2nd and 3rd division stages of the oocyte, and a nodal cell. In all instances, as well as could be determined, the chromosome number seemed to be from 12-14 (Plate 19, figs. 7-9). If the counts have been accurately made, the results of this study do not substantiate the contentions of Tuttle for his charophyte, that reduction occurs during gametogenesis; on the other hand, they are in accordance with the findings of Debski, Lindenbein, Oehlkers and others, that no reduction of chromosome number is found during gametogenesis.

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SUMMARY

The life history of a species of *Nitella* has been followed out as fully as possible under the circumstances, with the following results:

1. The plant has been determined as *Nitella hyalina* Agardh. As far as known, this is the first record of its collection in North America by an American collector.
2. A detailed description of the species is given.
3. A short description of the vegetative structure of the plant is presented in which are described the rotating nuclei of internodes and laterals.
4. Examples of vegetative reproduction are given.
5. In connection with the development of the antheridium:
 - (a) The apical cell is found to cut off only one basal cell at the beginning of the development, but later a second cell is cut off from the first, about the time of the 24-celled stage.
 - (b) A brief description of development of the antherozoids is presented.

- (c) The sperm are thought to escape by the perforation rather than by the gelatinisation of the mother cell walls.
- 6. In connection with the development of the oogonium:
 - (a) Several terms that have been given different meanings by Braun, Allen, Tuttle, Sachs, and others are defined and their interpretations as employed in the present paper set forth.
 - (b) An attempt is made to produce evidence that the three cells of the oocyte are cut off by successive divisions. This is contradictory to the findings of Tuttle in his investigations of a Charlottesville charophyte.
 - (c) The statement is made that the use of Braun's term "Wendungszellen" as meaning "wandering cells" or "turning cells" is not applicable to the species under observation, since the cells do not migrate from their original position, but merely appear to do so, being pushed aside by the growing oosphere.
 - (d) The number of stalk cells is found to be two instead of one as is usually stated for other species of *Nitella* and *Chara*.
 - (e) The first two divisions of the oocyte are shown to be more nearly oblique than vertical as is stated for other species of the Charales.
 - (f) A mitotic figure is presented to show that the lower tier of coronal cells is cut off from the sheath cell rather than from the first tier, as stated by Groves and Bullock-Webster.
- 7. A young plant bearing fruit and still attached to its oospore shell was found in a new aquarium about a month and a half after the first fruit was formed, seeming to indicate that the new zygote germinated without a previous long rest period.
- 8. A comparative study was made of the different types of nuclei found in different cells of the plant, revealing a wide range in shape, position, density, and in size from 7-600 μ .
- 9. A study of the deeply staining strands in the sheath cells was undertaken, and the conclusion reached that they are the nuclei of the cells and not bands of cytoplasm as thought by Groves and Bullock-Webster.
- 10. Chromosome counts were made for the following cells: sheath,

antheridial filament, coronal, 2nd and 3rd divisions of the oocyte. In each case, as well as could be determined, the number of chromosomes was found to be from 12 to 14. The results of this study do not substantiate the contentions of Tuttle for his charophyte, but are in accordance with the findings of Debski, Lindenbein, Oehlkers, and others that no reduction of chromosome number is found during gametogenesis.

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BIBLIOGRAPHY

1. ALLEN, T. F.
1880 The Characeae of America. Parts I and II. Boston. S. E. Cassino.
2. 1888 The Characeae of America. Part I. New York. Pub. by the author.
3. 1892-1896 The Characeae of America. Part II, fasc. 1-3. New York. Pub. by the author.
4. BAILEY, CHARLES
1882 The Structure of the Characeae. Manchester City News.
5. BRAUN, A.
1852 Ueber die Richtungsverhältnisse der Saftströme in den Zellen der Characeen. Monatsber. K. Akad. Wissensch. Berlin. 220-268.
6. DE BARY, A.
1875 Die Keimungs-geschichte der Charen. Bot. Zeit. 33: 409-419.
7. DEBSKI, B.
1897 Beobachtungen über Kernteilung bei Chara fragilis. Jahrb. wiss. Bot. 30: 227-48.
8. DREW, K. M.
1924 An abnormal pro-embryonic branch of Chara vulgaris L. Ann. Bot. 38: 207-9.
9. 1926 The leaf of Nitella opaca Ag. and adventitious branch development from it. Ann. Bot. 40: 321-48.
10. FRITSCH, F. E.
1935 The Structure and Reproduction of the Algae. London. Cambridge Univ. Press.
11. GOEBEL, K.
1918 Zur Organographie der Characeen. Flora 110: 344-87.
12. 1930 Die Deutung der Characeen-Antheridien. Ein Versuch. Flora 124: 491-8.
13. GOETZ, G.
1899 Ueber die Entwickelyng der Eiknospe bei den Characeen. Bot. Zeit. 68: 1-13.
14. GROVES, J., & BULLOCK-WEBSTER, G. R.
1920 The British Charophyta. Vol. I. London. Adlard and Son and West Newman, Ltd.
15. HALSTED, B. D.
1879 Classification and description of the American Species of Characeae. Proc. Boston Soc. Nat. Hist. 20: 169-190.

16. **HY, F. C.**
1913 *Les Characées de France.* Bull. Soc. Bot. France. Mem. 26: 19.
17. **KARLING, J. S.**
1926 *Nuclear and Cell Division in Nitella and Chara.* Bull. Torrey Bot. Club 53: 319-79.
18. 1927 *Variations in the Mature Antheridium of the Characeae.* Bull. Torrey Bot. Club 54: 187-230.
19. 1928 *Nuclear and Cell Division in the Antheridial Filaments of the Characeae.* Bull. Torrey Bot. Club 55: 11-39.
20. **LINDENBEIN, W.**
1927 *Beitrag zur Cytologie der Charales.* Planta 4: 437-66.
21. **McNICOL, M.**
1907 *The Bulbils and Proembryo of Lamprothamnus alopecuroides A. Braun.* Ann. Bot. 21: 61-70.
22. **MOTTIER, D. M.**
1904 *The Development of the Spermatozoid in Chara.* Ann. Bot. 18: 246-54.
23. **OEHLKERS, J.**
1916 *Beitrag zur Kenntnis der Kernteilungen bei den Characeen.* Ber. Deutsch. Bot. Ges. 34: 223-6.
24. **OLTMANN, F.**
1922 *Morphologie und Biologie der Algen, I.* 2nd edit. Jena.
- 24a. **PAL, B. P.**
1932 *Burmese Charophyta.* Journ. Linn. Soc. London 49: 47-92.
25. **ROBINSON, C. B.**
1906 *The Characeae of North America.* New York.
26. **ROZE, E.**
1867 *Lez antherozoides des cryptogames.* Ann. d. Sci. Nat. Bot. 5, 7: 87-103.
27. **SACHS, J. A.**
1882 *A Textbook of Botany, Morphological and Physiological.* 292-305. Oxford.
28. **TOMASI, J. A.**
1936 *Improving the Technic of the Feulgen Stain.* Stain Technology 11: 137-144.
29. **TUTTLE, A. H.**
1924 *The Reproductive Cycle of the Characeae.* Science 60: 412-413.
30. 1926 *The Location of the Reduction Division in a Charophyte.* Univ. Calif. Publ. Bot. 13: 227-34.
31. **WALTERS, E.**
1929 *Entwicklungsgeschichtliche und Cytologische Untersuchungen an einigen Nitellen.* Arch. Jul. Klaus-Stiftung 4: 23-121.

PLATE 16

PHOTOGRAPH OF SECTION OF PLANT SHOWING HABIT AND SEX ORGANS

, PLATE 17

STAGES IN THE DEVELOPMENT OF THE ANTHERIDIUM

Fig. 1. Very early stage showing apical cell (a.c.) and first basal cell (f.b.c.) resting on nodal cell of the leaf. X244.

- Fig. 2. Later stage in which a leaf primordium (l.p.) has been cut off from the node cell (n.c.). Note that the antheridium still has only one basal cell. Lowest cell in this row is a young internodal cell (i.c.). Note also that the cytoplasm of this cell is quite vacuolate. $\times 244$.
- Fig. 3. Still later stage, showing first vertical division of the apical cell, appearance of a leaf tip (l.t.) on one of the leaves, and of chloroplasts in the internode below the node. $\times 244$.
- Fig. 4. Octant stage produced by the transverse division. $\times 244$.
- Fig. 5. The 24-celled stage, also showing the appearance of the second basal cell (s.b.c.), just above the node. $\times 244$.
- Fig. 6. Section view of whole mount antheridium stained by the Feulgen technic, showing four shield cells with nuclei, four manubria with capitula and young filament nuclei. $\times 36$.
- Fig. 7. Mature antheridium, showing infoldings of shield cells. $\times 65$.
- Figs. 8 to 14 are figures from the development of the sperm. All $\times 480$.
- Fig. 8. Very young filament cells with their nuclei.
- Fig. 9. Late anaphase in two adjacent filament cells in the formation of four new consecutive cells.
- Fig. 10. Sperm mother cell nuclei.
- Fig. 11. Elongation of mother cell nuclei into incomplete bands. Note flagella.
- Fig. 12. Nearly mature sperm with flagella clearly seen.
- Fig. 13. Sperm body apparently within cell, with one flagellum free.
- Fig. 14. Mature sperm.
- Fig. 15. Manubrium with its single nucleus. $\times 409$.
- Fig. 16. Showing normal attachment of filaments to the secondary capitula (s.c.). $\times 409$.
- Fig. 17. Capulum with five filaments attached. $\times 409$.
- Fig. 18. A sketch to show the unusual attachment of filaments directly to the primary capitulum (p.c.). $\times 409$.

PLATE 18

STAGES IN THE DEVELOPMENT OF THE OOGONIUM

- Fig. 1. Initial row, showing in order from apex to base, (1) apical cell (a.c.) or primary oocyte, (2) nodal cell (n.c.), (3) and (4), two basal cells (f.b.c. = first basal cell; s.b.c. = second basal cell). $\times 244$.
- Fig. 2. Cutting off of first (1) of three little cells by primary oocyte. Also, appearance of a sheath primordium (s.p.) from the node. $\times 244$.
- Fig. 3. Stage showing anaphase in division of the oocyte nucleus to form the second little cell, whose nucleus will be formed from the basal group of chromosomes. $\times 244$.
- Fig. 4. Later stage, showing completion of the division in fig. 3, and the appearance of other sheath primordia. $\times 244$.
- Fig. 5. Much later stage showing the third mitotic division of the oocyte forming the third cell (3) (basal chromosome group) and egg or oosphere (upper chromosome group). $\times 244$.
- Fig. 6. Completion of divisions in the formation of the three little cells (1, 2, 3). Partial face view. $\times 244$.
- Fig. 7. Side view of fig. 6, showing displacement of the three cells due to the growth of the egg. $\times 244$.

- Fig. 8. Mature oogonium just prior to or following fertilization. Note hour-glass chamber and slits. $\times 86$.
Fig. 8a. Cross-section of a single spiral thread, showing the dark nucleus embedded in granular cytoplasm. $\times 244$.
Fig. 9. Spherical nuclei of young sheath primordia. $\times 244$.
Fig. 10. Nuclei elongation with cells. $\times 244$.
Fig. 11. Later stage of figs. 9 and 10, showing mature oogonium with greatly elongated sheath cell nuclei. $\times 88$.
Fig. 12. Mitotic stage in formation of coronal cells. $\times 244$.

PLATE 19

- Fig. 1. Very young leaf showing uninucleate tip and lower cell. $\times 244$.
Fig. 2. Older leaf with numerous nuclei produced by amitotic divisions. $\times 88$.
Fig. 3. Enlarged portion of edge of leaf to show cell wall, stationary chloroplasts, and rotating nuclei. $\times 409$.
Fig. 4. Young plant showing attachment of oospore case, rhizoids, the verticil of young leaves, and the two-jointed portion of the protonema above the verticil. $\times 23$.
Fig. 5. Abnormal oogonium on a long stalk. $\times 88$.
Fig. 6. Abnormal development of sheath cells from the base of a normal antheridium. $\times 88$.
Fig. 7. Showing chromosome counts in metaphase of antheridial filament cell. $\times 480$.
Fig. 8. Division of nodal cell to form a lateral.
Fig. 9. Enlarged view of fig. 8 to show late anaphase in mitosis of a vegetative division. $\times 409$.
Figs. 10 to 19 show a series of comparative sizes, shapes and densities of nuclei from various cells: $\times 409$.
Fig. 10. Lateral node cell.
Fig. 11. Young internode.
Fig. 12. Older internode.
Fig. 13. Sheath cell.
Fig. 14. Antheridial basal cell.
Fig. 15. Antheridial shield cell.
Fig. 16. Young leaf.
Fig. 17. Young and older spermatogenous filaments.
Fig. 18. Old leaf.
Fig. 19. Stalk cells of oogonium.
Fig. 20. Showing arrangement of chloroplasts in rows. Note line of "indifference" near lower center of figure. $\times 244$.

PLATE 16



PLATE 17

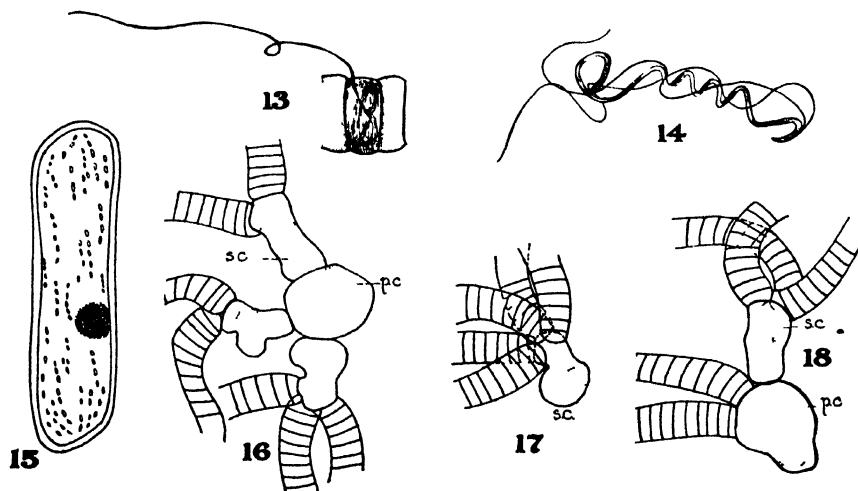
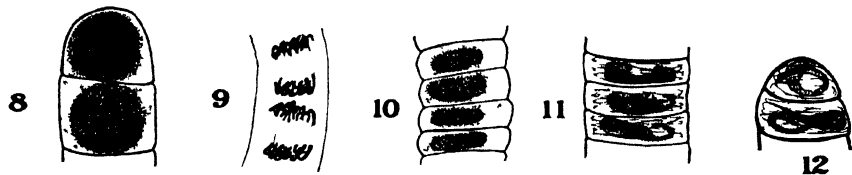
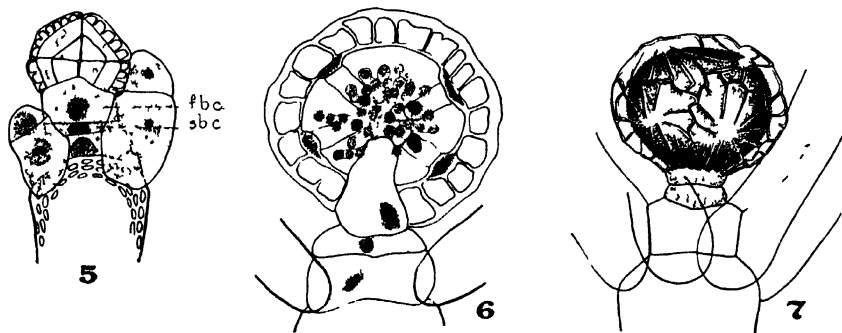
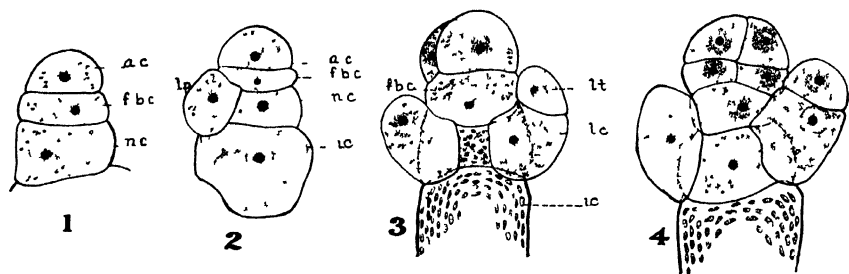


PLATE 18

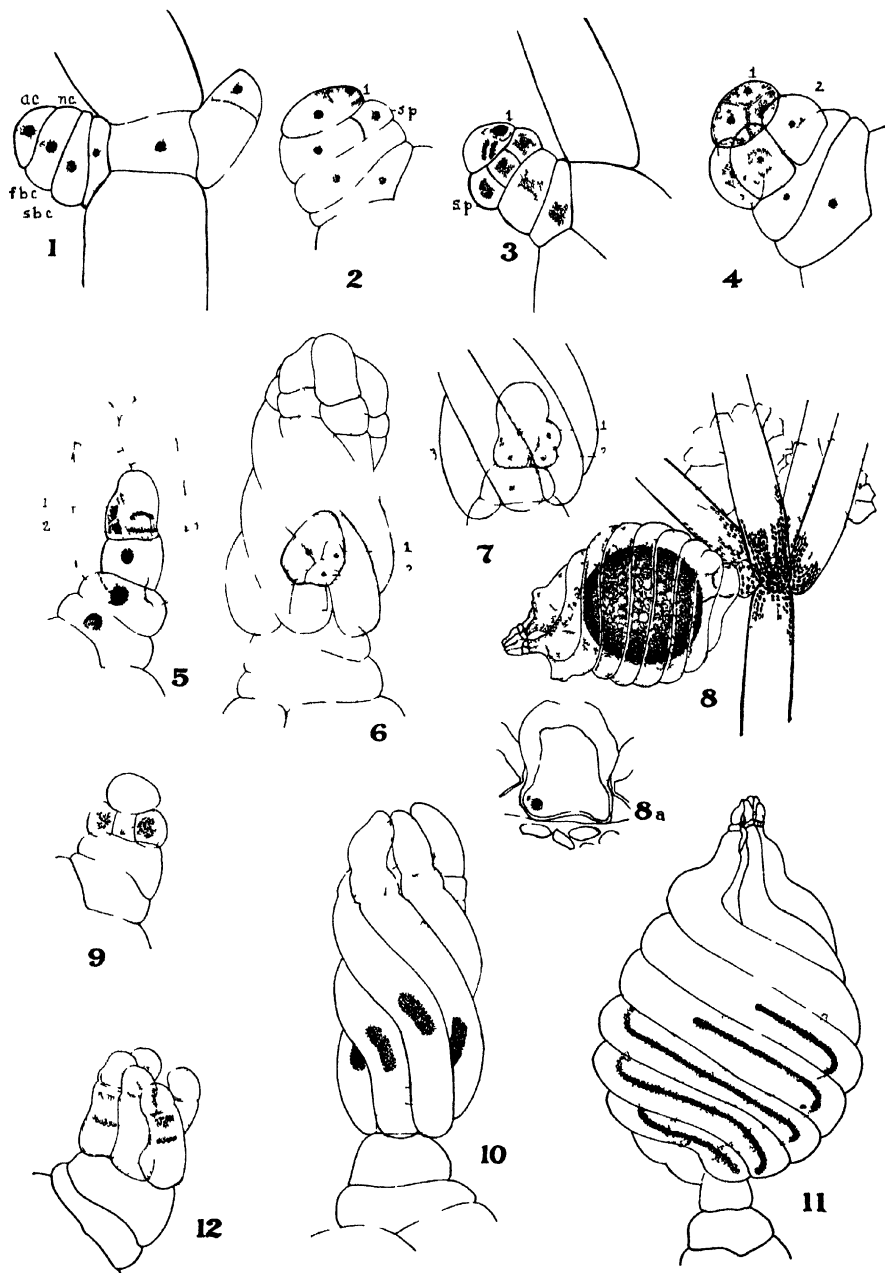
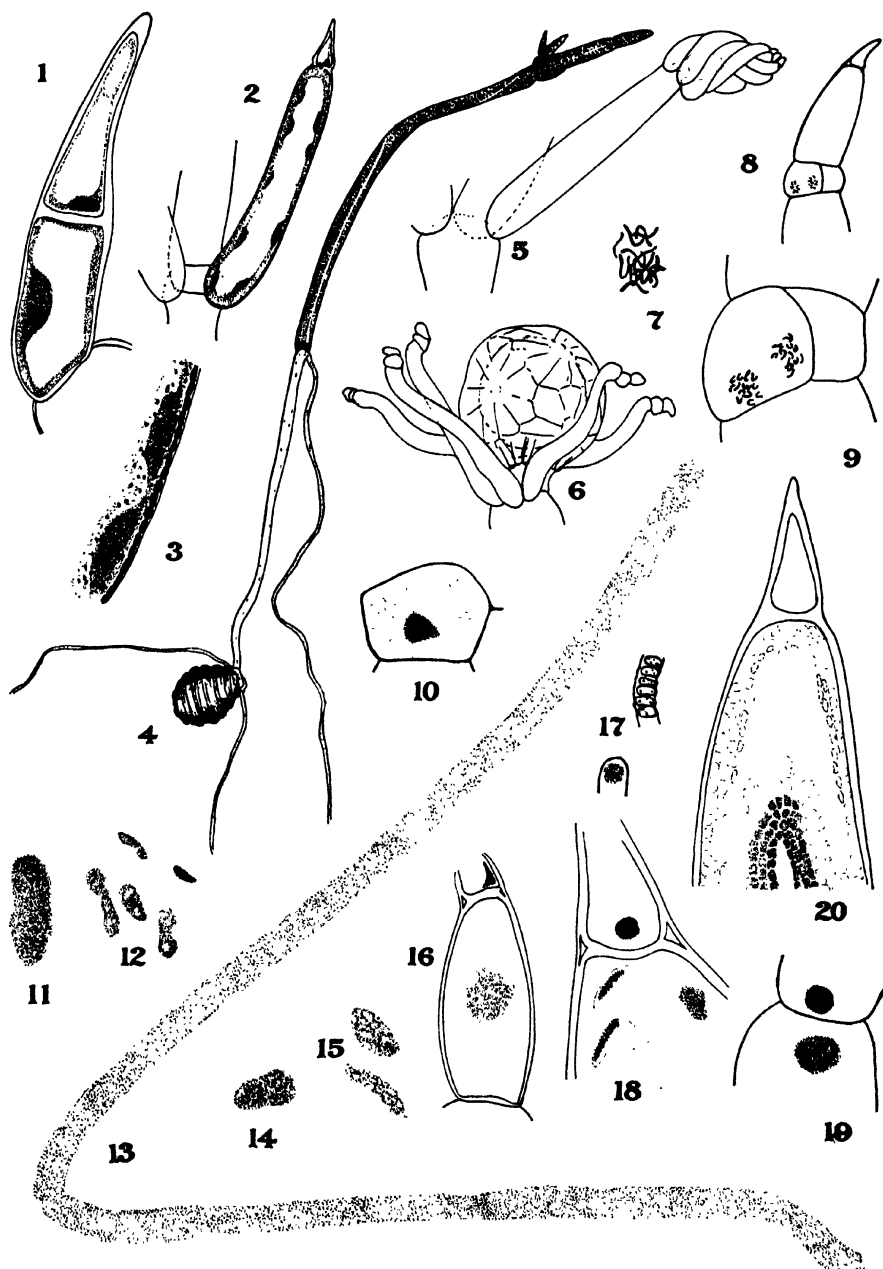


PLATE 19



A NEW GENUS OF THE BLASTOCLADIACEAE

By VELMA D. MATTHEWS

PLATES 20 AND 21 AND ONE TEXT FIGURE

During the summer of 1935 the writer found a most unusual fungus growing on a small dead fly in a pool in a depression of a large rock on Bald Knob in Giles County, Virginia. The elevation at this point is about 4,200 feet. At this time only a few plants were observed and all attempts to culture the fungus proved in vain. However, in the summer of 1936 the same fungus was found growing on an insect in the same pool. This time the writer succeeded in growing the fungus on house fly eggs, which had just been removed from a fly and placed in sterile water. If the fly eggs were of the proper maturity an abundant group of plants usually grew on at least a few of the eggs, when they were placed in a growing culture. During the winter, when fly eggs were more difficult to obtain, cultures were made on the radicle of hemp seed embryos which had been boiled a few minutes. In these cultures the plants were often larger than those on fly eggs. The cultures have been kept in the refrigerator except when observations were being made. Germination of the spores was obtained on corn meal dextrose agar. On this medium the plants were small and short and usually produced a resting sporangium. However, young plants may be flooded with water and be made to develop thin-walled sporangia and zoospores.

The entire plant is made up of a sporangium or a resting sporangium, a main trunk, and a system of rhizoids. Plants bearing resting sporangia resemble a miniature *Allomyces* or *Blastocladia*. However, the main trunk never branches, so far as I have been able to discover. The zoospores, which are produced in large numbers in sporangia placed in fresh water are elliptic to oval with one long posterior cilium. They escape fully formed into a small vesicle, which persists only a few seconds or directly into the water. Here they swim for varying lengths of time and come to rest as round spores with a definite wall. The swimming spore has the same type of structure as the spores of *Allomyces* and *Blastocladia*. Upon coming to rest on a suitable sub-

stratum the spores send out a slender tube, which may soon fork and these branches divide up to form an elaborate system of very fine rhizoids, which extend into the substratum. The original spore body at the same time enlarges and develops into the trunk and sporangium. No reaction was obtained with chloriodide of zinc on the young or old plants. (*Dictyuchus* threads tested at the same time gave a beautiful purple reaction.)

The sporangia are thin-walled and globose to cylindrical in shape. Variation in size is very great, apparently depending on the amount of food material available. The method of the formation of the zoospores is shown in figures 2 to 8. The cytoplasm with many large refractive granules collects in the tips of the young plant, leaving a highly vacuolated basal portion (fig. 2). After most of the cytoplasm and granules have collected in the tip a wall begins from the periphery and finally makes a complete partition separating the tip from the remainder of the plant (fig. 3). Some enlargement of the sporangium now takes place and the refractive granules become smaller and scattered throughout the very fine cytoplasm (fig. 4). A few minutes after this stage these granules are much smaller and arranged in a more or less circular manner (fig. 5). A few minutes later the origins of the individual spores become visible with a group of granules in each. Five to ten minutes later the conspicuous papilla suddenly seems to be pulled inward, movement of the spores begins, and the papilla is pushed outward to form a very thin vesicle, which lasts only a few seconds. No cap is cut off. After the vesicle bursts the spores separate and swim away with their cilia propelling them from behind. The zoospores remaining in the sporangium come out rather rapidly and swim away. The structure of these spores is similar to that described by Hatch (2) in gametes of *Allomyces* and by Cotner (1) for the zoospores of *Blastocladia*.

Some plants in all large cultures form resting sporangia in place of the ordinary thin-walled sporangia. In cultures where fresh water is not added, practically every plant forms a resting sporangium. The tip of the plant is separated off by a cross wall and inside of this tip the protoplasm assumes a brownish color, which gradually deepens and a wall is formed around the entire contents (fig. 15). This wall becomes thick and irregularly marked on the outer surface (fig. 16). After a rest of two to three weeks, in some cases perhaps less, the germination of some of the resting sporangia can be obtained by adding fresh water and food material to the culture. Twenty-four hours to

ten days after the addition of fresh water and food the heavy wall of the sporangium cracks in several places and one to three thin-walled exit tubes appear. The exit tubes are usually less than the diameter of the sporangium but in a few cases very long ones developed as shown in figure 19. The outer hyaline wall, which was the original wall of the hyphal tip may or may not be present at the time of the germi-



Culture of *B. simplex* growing on a fly egg. Note three large plants bearing resting sporangia and many small sporangia. $\times 80$

nation of the resting sporangium. When the sporangia are three or four months old there is usually no indication of this wall but the resting sporangia are still in contact with the old trunk. Soon after the appearance of the exit tubes the contents of the resting sporangium become divided up into spores similar to those found in the ordinary sporangia. The first of the spores escape into a very thin vesicle

lasting only a few seconds or directly into the water. Hundreds of spores are formed in the large resting sporangia so several hours may be necessary for their escape. These zoospores germinate to form plants exactly like those formed by the zoospores from the thin-walled sporangia. There is no inherent alternation between the thin-walled and the thick-walled sporangia, their occurrence depending on environmental conditions. Mature resting sporangia may be gradually dried by allowing the water in the petri dish (with top on) to evaporate in the refrigerator or outside the refrigerator, when the temperature is not above 70°F. In the dried condition sporangia were kept 4 months and germination obtained.

A new genus *Blastocladiella* is suggested to include the plant described above.

***Blastocladiella* gen. nov.**

Minute exposed part a simple thin-walled trunk without constrictions bearing at its tip a thin-walled sporangium or a thin case containing a thick-walled resting sporangium. Zoospores with one posterior cilium, monoplanetic. Rhizoids delicate, much branched. Sexual reproduction unknown. Walls not turning blue in chloriodide of zinc.

***Blastocladiella simplex* sp. nov.**

Trunk 8–40 μ broad and 30–1,005 μ long (even shorter on agar, fig. 12), without constrictions and unbranched except at base, where it may divide into 2 to 4 parts from which the system of very fine rhizoids extends. Sporangia cylindrical to globose, 15–105 μ in diameter, usually with one, rarely up to three papillae of emergence. Zoospores monoplanetic, oval to elliptic, 3–4 x 5.5–7 μ , with a long posterior cilium, a nucleus with a large nuclear cap, a ring of glistening granules, a large vacuole and very fine cytoplasm. Resting sporangia 15–180 μ in diameter formed inside a wall shaped as the thin-walled sporangia and developing a brown irregularly marked heavy wall. The thin-walled case does not dehisce to allow the escape of the sporangium, but it may disappear in old cultures. After a rest the thick-walled sporangia form zoospores similar to those formed in the thin-walled sporangia. Sexual reproduction unknown.

Growing on a dead fly in a fresh water pool on Bald Knob near Mountain Lake, Giles County, Virginia.

In general appearance *Blastocladiella* resembles *Macrochytrium botry-*

dioides Minden of the Chytridiales, but the absence of a cap and the structure of the spores lead us to place our plant in the Blastocladales.

Blastocladiella may be separated from *Blastocladia* and *Allomyces* by the unbranched trunk and the formation of only one sporangium on a plant and from *Allomyces* and some species of *Blastocladia* by the resting sporangium not escaping from the thin outer wall. The presence of a vesicle also separates the new genus from *Blastocladia*. *Blastocladiella* in its simplicity may be considered as a connecting link between the Chytridiales and Blastocladales.

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HARTSVILLE, S. C.

LITERATURE CITED

1. COTNER, F. B.
1930 Cytological Study of the Zoospores of *Blastocladia*. Bot. Gaz. 89: 295.
2. HATCH, W. R.
1935 Gametogenesis in *Allomyces arbuscula*. Ann. Bot. 49: 623.
3. MINDEN, M. v.
1915 Kryptogamenflora der Mark Brandenburg 5: 385.

EXPLANATION OF PLATES 20 AND 21

PLATE 20

- Fig. 1. Habit sketch of a culture growing on a fly egg. $\times 53$.
Figs. 2-8. Stages in the development of a sporangium. Fig. 2 drawn at 10:55 A.M.; fig. 3, 11:20 A.M.; fig. 4, 1:00 P.M.; fig. 5, 1:45 P.M.; fig. 6, 2:09 P.M.; fig. 7, 2:10 P.M.; fig. 8, 2:30 P.M. All $\times 460$.

PLATE 21

- Fig. 9. Zoospores from a thin-walled sporangium. $\times 1320$.
Fig. 10. Germinating zoospores. $\times 460$.
Figs. 11-12. Young plants grown on agar. $\times 111$.
Figs. 13-14. Entire plants with short trunks and thin-walled sporangia. $\times 220$.
Fig. 15. Young resting sporangium. $\times 460$.
Fig. 16. Mature resting sporangium. Outer wall has disappeared. $\times 460$.
Fig. 17. Resting sporangium forming an emergence tube. $\times 460$.
Fig. 18. Resting sporangium with zoospores escaping into a vesicle. $\times 460$.
Fig. 19. Small resting sporangium with a very long emergence tube. $\times 460$.
Fig. 20. Zoospore from resting sporangium. $\times 1320$.
Fig. 21. Very small sporangium stained to show nuclei in young spores. $\times 1320$.

PLATE 20

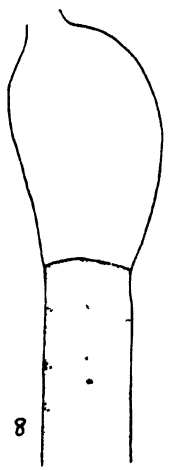
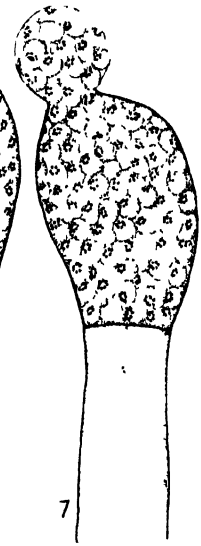
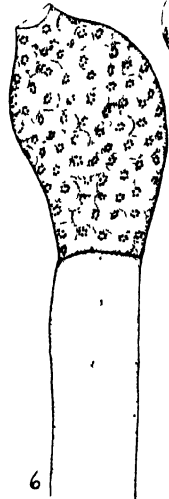
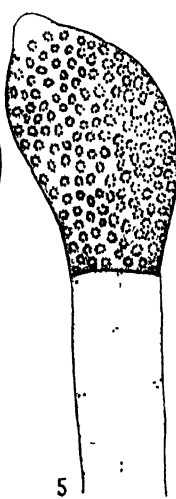
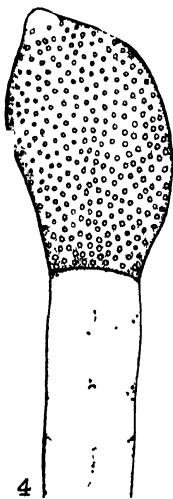
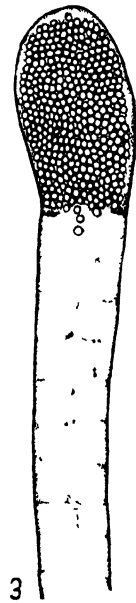
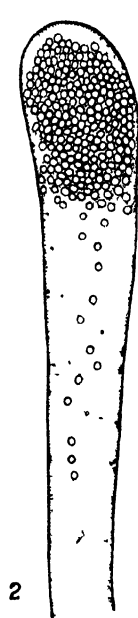
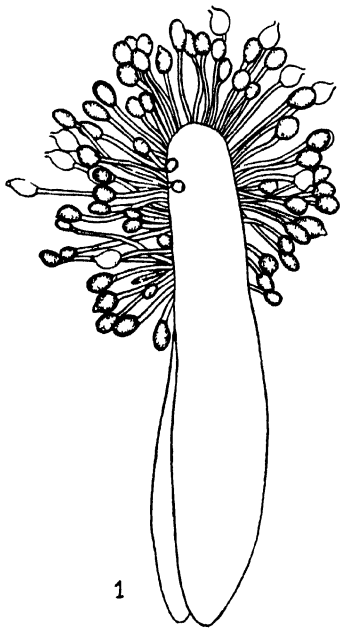
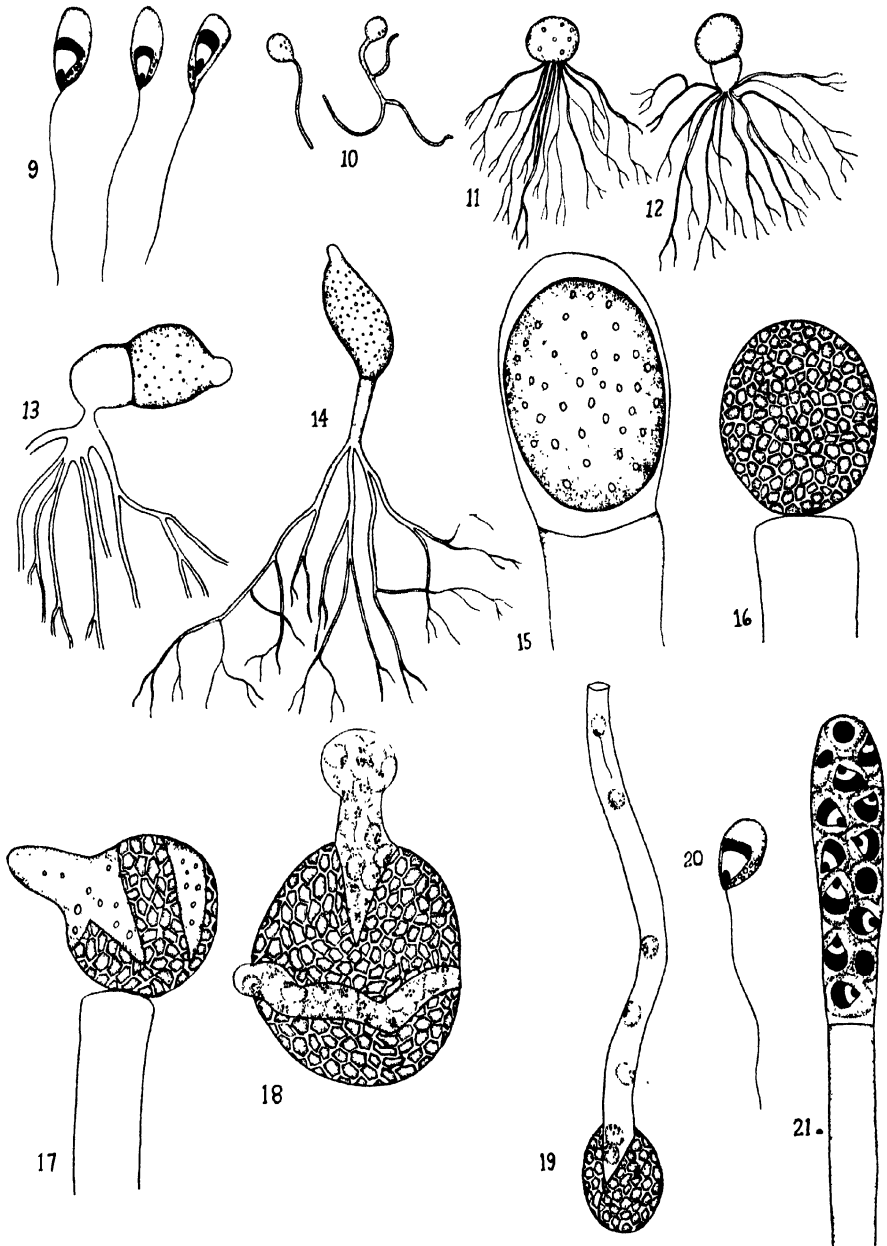


PLATE 21



A NEW SPECIES OF ROZELLA PARASITIC ON ALLOMYCES

By FRANCES K. FOUST

PLATES 22 AND 23

A new species of *Rozella* was found on *Allomyces arbuscula* Butler in a soil collection from Battle Grove, across the road from Coker Arboretum, Chapel Hill, N. C., October 5, 1936. This organism is not only a new species but is the first recorded parasite on the genus *Allomyces*. However its greatest significance lies in the fact that through studying the parasite in the laboratory several points have been cleared up which have long been the basis of controversy. Cornu (1871), working on *Rozella septigena*, observed the spiny resting bodies associated with the parasite when growing on members of the Saprolegniales, but was unable to determine whether the bodies belonged to the parasite or to the host. He observed the formation and discharge of the zoospores in *Rozella Rhizidi spinosi*; but he stated that they "decompose without germination: one wonders if they are really zoospores." Others have worked on *Rozella* since Cornu and have reported the spiny resting bodies. They have been unable, however, to observe the germination of these resting bodies and thus give definite proof that they belong to the parasite. The number and place of attachment of the cilia has also been a disputed point.

This investigation has been carried out under the direction of Professor J. N. Couch, to whom the writer tenders her sincere thanks.

The parasite was isolated and successfully kept in culture by the use of the following method. A non-parasitized strain of *Allomyces arbuscula* already in culture in the laboratory was purified in agar and used as a stock culture for inoculation with the parasitized material. Young *Allomyces* cultures on hemp-seed were inoculated (before host resting bodies had begun to form) by placing a few of the parasitized host threads containing resting bodies in direct contact with the host filaments. In five to twenty-four hours after inoculation (depending upon temperature), the *Rozella* sporangia can be found in the early stages of development at the hyphal tips of the host. The life cycle is completed in from two to three days with the formation of the resting bodies, which retain their vitality over a period of several months.

Rozella allomyces n. sp.

Fungus body parasitic within the distal parts of the threads of *Allomyces*. Sporangia first formed at the tips of the young host threads, usually 1-5 in a row, in basipetal succession; usually barrel-shaped, but varying greatly in size and shape, $12-20 \times 20-40\mu$, more often about $15.9\mu \times 24.6\mu$. The wall apparently confined to the original host wall. Usually with one exit papilla, which is about 1.3μ long. Frequently a primary sporangium may be divided by one or more partitions into several smaller sporangia. Zoospores ovoid, the posterior end the larger, about $3-4\mu$ thick, containing one refractive globule and with one posteriorly attached cilium, which is four times the length of the spore and is directed backward when the spore swims. Swimming by darting about with frequent changes of direction as typical for Chytrid spores. The swimming period lasting for about an hour, after which the spores germinate or die. Resting bodies formed later than the sporangia, occurring in the distal part of the host threads just behind the sporangia in swollen segments that are 1-35 in number, each segment containing from 1-16 resting bodies, the average being about 3 or 4. Segments spherical, barrel-shaped, nearly cylindrical, or irregular, $20-40 \times 20-70\mu$. Segments not completely filled by resting bodies, usually containing some left-over, dead, granular, host protoplasm. Resting bodies with spiny walls, spherical, $12-20\mu$ thick, averaging 15.9μ thick (average of 20 measurements) counting the spines which are about 1.3μ long; yellowish brown to reddish brown in color; when mature with a thick ($.5\mu$) wall, with a central hyaline globose mass of material surrounded by a granular zone of protoplasm. Germinating after a rest period of a week to form zoospores if fresh water is added and a young culture of *Allomyces* is present. The resting bodies may retain their vitality either dry or wet for several weeks, perhaps months. Zoospores from resting bodies identical with those from regular sporangia and capable of infecting young host threads.

SPORES AND SPORANGIA

Spores have been observed to swim for about an hour before coming to rest. They apparently do not pitch on trash or other nearby objects but only on the filaments or reproductive bodies of the host. The primary infection seems always to take place at the tip of the host thread. The spore loses its cilium, encysts and sends a fine tube through the host membrane (figs. 16-20). The protoplasm in the spore then flows through this tube into the host. In mature infected cultures the spores may settle on the older parts of the host threads and successfully penetrate the host wall. In such cases the host protoplasm forms callosities which wall off the invading parasitic bodies and thus prevent their further development (figs. 18, 19). This probably explains why infection is unsuccessful with old cultures of *Allomyces* and with older parts of threads in younger culture.

In early stages of development it is impossible to decide just how much of the protoplasm in an infected hyphal tip belongs to the parasite and how much to the host. The parasitic sporangia are usually formed in those tips that have not begun noticeable development into sporangia or resting bodies. However, resting bodies of the host in all stages of maturity have been found which were parasitized and later produced *Rozella* spores. The parasitic protoplasm becomes more or less evenly scattered among the larger granules of the host cytoplasm. Meantime the infected hyphal tip has a pale whitish gleam which contrasts strikingly with the darker and more coarsely granulated protoplasm of the host. A septation is formed cutting off the parasitic sporangium from the rest of the hypha. If a separate wall is laid down by the parasite to enclose the sporangium it is so thin and closely applied to the host wall as to be indistinguishable as a separate membrane. Sometimes one or more partitions are laid down within the primary segment. The partitions may be horizontal or vertical or placed at such angles as to appear Y-shaped in section. The formation of such a cross wall takes place in this manner: a clear furrow is seen to extend across the cell, widening out at the sides where it joins the cell walls. Later small granules are deposited in the furrow and these appear to fuse to form the partition.

Soon after the cross wall has been formed, one or more vacuoles are seen actively vibrating. The protoplasm around the vacuoles is composed of the heavy coarse granules of the host and the fine clear granules of the parasite. Apparently small clumps of the host plasma are digested in the vacuoles. As the sporangium matures the vacuoles are seen to fuse so as eventually to form one or more large vacuoles which become centrally located. The spore origins are cut out and move gently, while the clear vacuolar spaces are still visible with the vigorously dancing granules. A disappearance stage occurs, during which the spore origins and vacuoles disappear, the granules becoming evenly dispersed in the homogeneous protoplasm. At this stage there seems to be a general swelling of the entire contents. Early in the development, about the time the cross wall is formed, a small protuberance is seen to appear on the outer wall of the sporangium; and as the development within the sporangium increases, the papilla enlarges.

A few minutes after the disappearance stage the granules begin to collect in small evenly spaced groups, each group forming the center of a spore (pl. 23, figs. 6, 7). Quickly the spores take on their form and a faint rocking and jerking motion becomes evident. As the movement increases in rapidity, the outlines of the spores become distinct. Soon

the spores are in a seething motion, moving in whirlpool-like currents (fig. 8). The walls continue to bulge out and the papilla to enlarge (figs. 9-11). The papilla wall becomes quite thickened, as though it were being gelatinized, and swells outward. Meantime the mass of spores is pushed inward by a clear hyaline space which is a continuation of that of the papilla. This space continues to push the spores backward; at the same time the bulge of the papilla is greatly increased. Suddenly the tip bursts and the spores are shot out in a straight stream (fig. 12). So great is the internal pressure that the spores and small granules of matter are shot out in a mass in which it is hardly possible to distinguish the spores until they are a little distance from the sporangium. Their motion is then slowed up, and it can be seen that the spores come out with their cilium directed backwards. They dart about very rapidly, frequently changing their direction. The majority of the spores have escaped from the sporangium in two or three minutes after the tip bursts. Occasionally some of the spores do not escape from the sporangium but dart rapidly about within the walls. In stained material spores have been seen within the sporangium which had two cilia and were twice the size of normal spores. These biciliate spores are probably due to incomplete segmentation in the sporangium. On one occasion spores were seen within a sporangium to creep about the wall in an amoeboid fashion. These however resumed their rapid motion and escaped if they happened to find the exit tube.

Spores of this genus were originally described by Cornu (1872) as being ovoid and typically uniciliate. However, Fischer (1894) describes the spores as possessing two cilia, a short one in front and a long one behind. We are very certain that, in the present species, the spores are uniciliate. They have been seen on numerous occasions and in exceedingly large quantities. One of the most favorable methods by which actively swimming spores can be observed is in a small drop of water on a slide without a cover slip in which a large number of spores are swimming in the edge of the drop. If such spores are observed under a high dry lens with correction collar, a single cilium can easily be made out. A single cilium has also been observed with a water immersion lens. Preparations killed with the fumes of osmic acid and stained with a weak solution of gentian violet and mounted in water show beyond doubt the presence of a single cilium (pl. 22, fig. 3). Spores stained by the Cotner method gave similar results. There can hardly be any doubt but that there are two groups of plants in this family, one having uniciliate spores and the other biciliate spores.

RESTING BODIES

The spiny resting bodies are formed late in the development of the culture after the *Rozella* sporangia have matured (figs. 21-26). They appear below the sporangia in the same filaments and in the same type of host segment. The segments are spherical, barrel-shaped or long cylindrical, depending on the size and number of resting bodies in one segment. As with the sporangia the resting bodies are formed in basipetal succession, the older being found in segments next to the youngest sporangia. All stages in the development of the resting bodies may frequently be seen in one filament. There seems to be a definite zone below which successful infection does not take place, the infected zone usually occupying from a third to a half of the distal part of a hypha. The first crop of spores from the first formed sporangia reinfect the host threads to form other sporangia. The second group of sporangia produce spores which may reinfect the host to form resting bodies or infect new hyphae to form sporangia. When conditions in the culture change, such as shortage of food supply, change in temperature, etc., the infecting plasmodia form resting bodies instead of sporangia. A somewhat analogous behavior has been found by Barrett (1912) to exist in *Olpidiopsis*, where sexual organs are produced when unfavorable conditions arise.

The plasmodium may be seen as a clump of dark granules some of which are large and coarse, while others are fine and lighter in color. Vacuoles become distinct in the host protoplasm surrounding the parasitic body, and these grow larger as the parasitic plasmodium becomes larger. The plasmodium rounds up, and a very delicate membrane is formed around it. A clear, hyaline space surrounds the body. Into this circular space granules are soon seen crowded close to the parasite wall, which has become thicker. These granules line up at right angles to the spore wall and appear to fuse forming the spines. The exact nature of the contents of the mature resting body can only be ascertained by cytological methods, which will be employed later. It seems, however, that the mature resting body contains a large globule of hyaline material surrounded by protoplasm. These bodies remain within the host thread until the walls of the host disintegrate.

Under the proper conditions or stimuli the resting bodies germinate (fig. 27). The consecutive development of a resting body has not been followed, but numerous stages in their development have been observed. It is possible to recognize a germinating resting body by the presence of the emergence papilla. In a relatively late stage of spore development

the contents of the sporangium become evenly dispersed throughout and begin a vigorous movement. After the spores are cut out, they begin a slow motion near the periphery of the body, which gradually increases in rapidity until the whole mass is seething. As the development of the contents continues, the papilla swells and enlarges. Suddenly the papilla bursts, and the spores are liberated in much the same manner as the spores from the primary sporangium and to all appearances are similar to the latter in structure and behavior. No fusion of spores seems to take place. If the host wall has not become broken down or torn the spores are liberated within the host filament, as was the case in most of the observed cultures.

Proof that the spiny resting bodies belong to the *Rozella* was obtained by reinfection experiments. Host threads containing spiny resting bodies (a week or more old) were cut off and placed in contact with the young hyphae from a pure strain of *Allomyces arbuscula*. After 24 hours time the resting bodies had germinated and many of the hyphal tips of the *Allomyces* were infected with the characteristic sporangia of the parasite, thus proving the position of the spiny resting bodies in the life history. None of the previous observers has been able to obtain the germination of these resting bodies and to demonstrate their positive relationship to *Rozella*.

SUMMARY

1. A new species of *Rozella* parasitic on *Allomyces arbuscula* is described.
2. The zoospores possess one posteriorly attached cilium which is directed backwards when swimming.
3. The germination of the spiny resting body has been observed a number of times. In the structure and mode of exit, spores from the resting body are similar to those from the primary zoosporangium. The identity of the resting bodies with the parasite has been definitely proved by reinfesting the healthy *Allomyces* cultures with the spores from germinating resting bodies.
4. No evidence of fusion of any spores has been found.

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LITERATURE CITED

BARNETT, J. T.

1912 Development and Sexuality of Some Species of Ovipidiopsis, (Cornu)
Kitcher. Ann. Bot. 26: 209-232, pls. 23-30.

CORNU, M.

1872 Monographie des Saprolegniées. Deuxième partie. Chytridiées parasites des Saprolegniées. Ann. Sci. Nat. Bot., ser. 5, 15: 112-198, pls. 3-7.

FISCHER, A.

1894 Ueber die Geisseln einiger Flagellaten. Pringsheim's Jahrb. f. Bot. 28: 187-235, pls. 11, 12.

EXPLANATION OF PLATES

PLATE 22

All photos except 3 from living material.

- FIG. 1 Part of a young culture of *Allomyces arbuscula* Butler about eighteen hours after infection showing hyphal tips infected by parasite, *Rozella* (p). Note pale, whitish gleam of young parasitic sporangia. Note also much larger and denser resting bodies of host (r.s.) $\times 120$.
- FIG. 2. Four heavily parasitized hyphal branches of *Allomyces*. Toward distal part of host thread, sporangia of parasite are formed, all of which have emptied except one. This one (s_1) shown in filament *b* discharged its spores a few minutes after photograph was made. The segment at tip of thread *b* is divided into three sporangia (s). The dark rounded bodies, of which one to four are shown in each segment, are the resting bodies of parasite. The older ones are toward hyphal tip, the younger stages nearer base. Note very early stage of infection in thread *a* at *i*. $\times 250$.
- FIG. 3 Two zoospores showing single cilium on each. $\times 1620$.
- FIGS 4-6. Enlarged views of threads *a*, *b*, *c*, respectively shown in fig. 2, showing stages in development of resting bodies. Note young spines on resting bodies in figs 5 and 6. $\times 540$.
- FIGS. 7 Resting bodies of *Rozella*. Note central globule, and spines. Two of the resting bodies have discharged their spores and partially collapsed $\times 1140$.

PLATE 23

- FIG. 1. Habit sketch of parasitized *Allomyces* thread, showing empty zoosporangia of parasite and resting bodies in various stages of development. Note young plasmodia of parasite in lower part of figure. $\times 187$.
- FIGS. 2-7. Stages in development of sporangia from just after infection to formation of spores. Fig. 4 shows large central vacuoles with spore origins (?). Fig. 5, disappearance stage. Fig. 6, reformation of spores. Fig. 7, spores ready to emerge. $\times 787$.
- FIGS. 8-12. Diagrammatic sketches showing spore discharge. See text for explanation. \times about 787.
- FIG. 13. Spores as they appear in the living condition. $\times 1050$.
- FIG. 14. Three primary segments which have become divided into two or three secondary segments, each a sporangium. Note exit pores. $\times 483$.
- FIG. 15. Biciliated zoospores which had failed to emerge from sporangium. $\times 980$.
- FIG. 16. Spore, still with cilium, on host thread. $\times 980$.

- FIGS. 17-20. Stages in infection.** Fig. 17, infection tubes starting. $\times 1046$.
Fig. 18. Two spores, one of which has sent germ tube through host wall and emptied most of its protoplast into host thread. The host thread is forming a callus around parasite and it is doubtful if this infection would have developed further. $\times 1312$. Fig. 19, another spore showing same condition as above. $\times 1312$. Fig. 20, collapsed membrane of spore which apparently has emptied its contents into host. Parasite plasmodium is shown by darker granules $\times 1312$.
FIGS 21-26. Development of resting body, for explanation see text. $\times 787$.
FIG. 27. Resting body which has germinated forming zoospores, all of which have been discharged except four $\times 787$.

PLATE 22

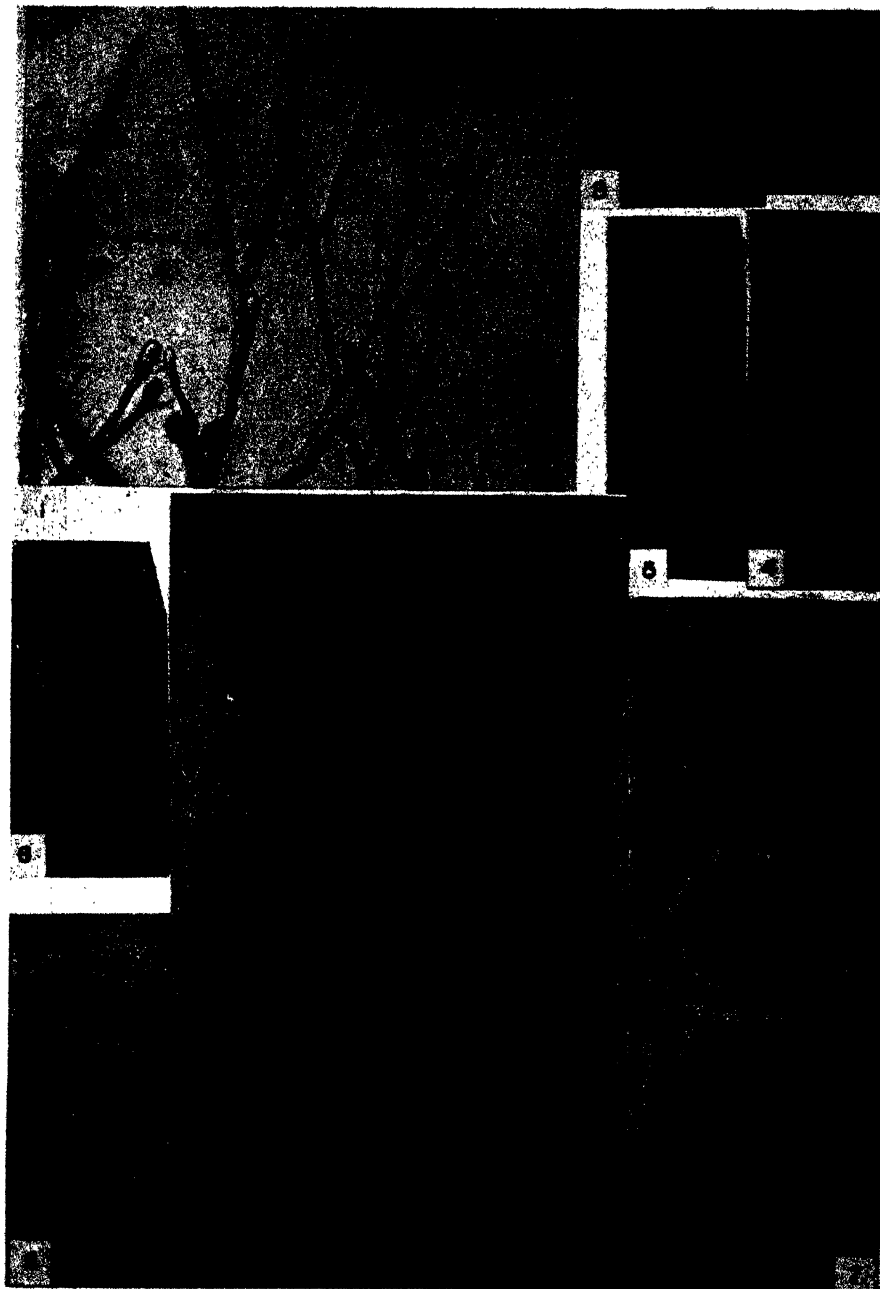
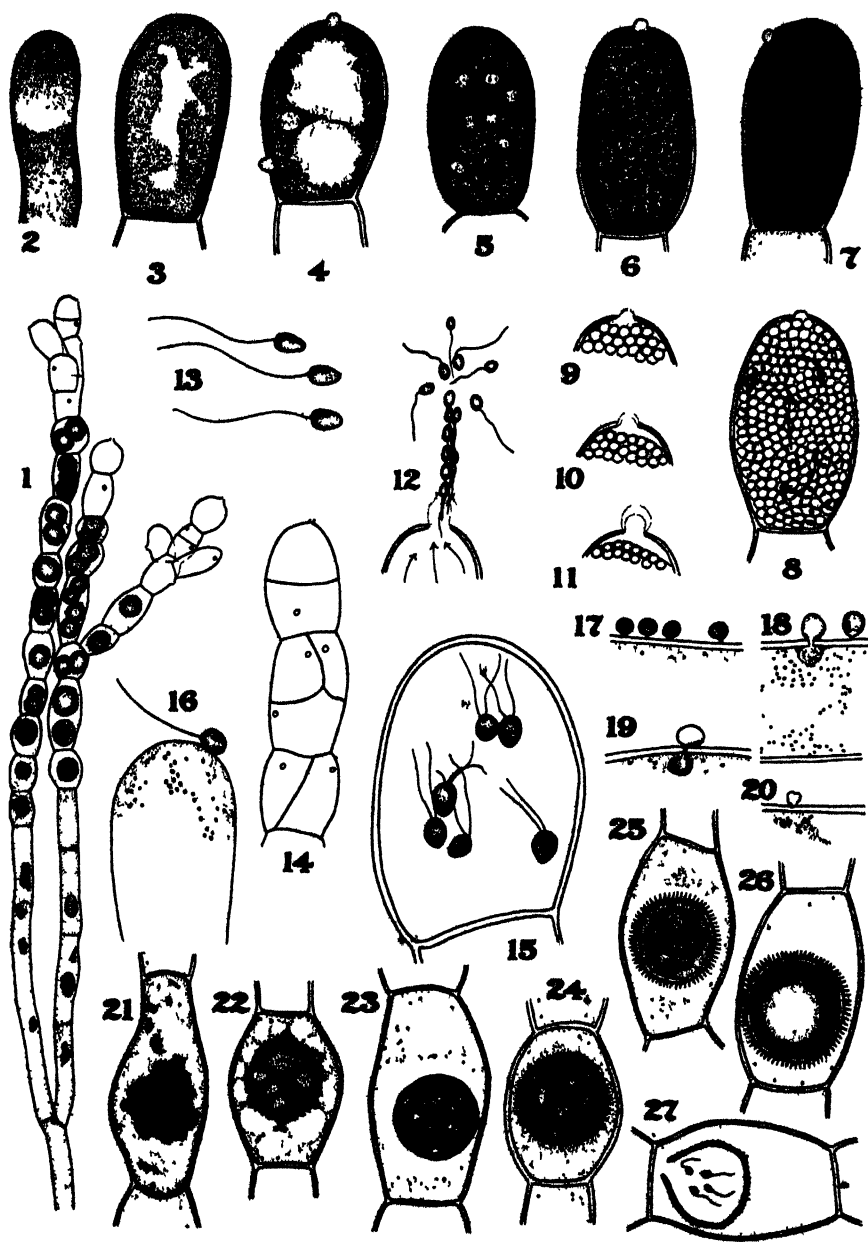


PLATE 23



JOURNAL
OF THE
Elisha Mitchell Scientific Society

Volume 53

December, 1937

No. 2

PROCEEDINGS OF THE THIRTY-SIXTH ANNUAL MEETING
OF THE NORTH CAROLINA ACADEMY OF SCIENCE

CATAWBA COLLEGE, SALISBURY, N. C., MAY 7 AND 8, 1937

The thirty-sixth annual meeting of the North Carolina Academy of Science was held at Catawba College, May 7 and 8, 1937. The meeting was called to order at 9:30 A.M. on May 7 by the President, P. M. Ginnings. The reading of papers commenced promptly and continued until 11:30 A.M. when the President announced the appointment of the following committees:

Auditing: J. B. Derieux, G. H. Satterfield, A. F. Thiel;

Resolutions: J. P. Givler, R. J. Campbell, E. E. Randolph;

Nominating: H. B. Arbuckle, J. S. Holmes, F. A. Wolf.

The reading of papers was continued until 1:30 P.M. when the Academy took a recess for luncheon.

The reading of papers was resumed at 2:15 and continued until 4:00 when the Academy held its annual business meeting.

The minutes of the previous meeting were approved as published in the JOURNAL OF THE ELISHA MITCHELL SCIENTIFIC SOCIETY.

Reports from the various committees were presented.

The executive committee, consisting of P. M. Ginnings, president of the Academy; C. F. Korstian, vice-president; H. L. Blomquist, secretary-treasurer; W. E. Speas, H. R. Totten, and W. L. Porter, reported as follows:

"The executive committee met in Salisbury on May 6 and on May 7 with all members present.

"The committee appointed W. L. Porter to act as temporary assistant to the secretary during the meeting.

"One title which had been accidentally omitted was added to the program.

"One title which arrived too late to go on the printed program was added to the program.

"Extension of time up to 30 minutes was granted to Frank K. Cameron for his paper 'The Utilization of Whole Cotton'.

"The request by G. R. McCarthy and J. W. Huddle that their paper be read in their absence by Grover Murray was granted.

"The secretary was instructed to see that in all the sections, except the chemistry section, no officer be elected who is not a member of the Academy in good standing. The committee expressed the desirability that the secretary of the chemistry section be a member of the Academy so that he may receive the announcements and the program of the Academy.

"The committee reported as elected to membership since the last meeting the following:

Dr. J. E. Adams, Dept. of Botany, U. N. C., Chapel Hill, N. C.

Mr. Bill Arey, 102 South Church St., Salisbury, N. C.

Mr. Walter Lane Barksdale, Dept. of Botany, U. N. C., Chapel Hill, N. C.

Dr. E. Willard Berry, Dept. of Geology, Duke University, Durham, N. C.

Dr. Robert B. Burrows, Box 221, Elon College, N. C.

Mr. George A. Christenberry, Dept. of Botany, U. N. C., Chapel Hill, N. C.

Mr. J. M. Clarkson, N. C. State College, Raleigh, N. C.

Miss Roxie Collie, N. C. State Museum, Raleigh, N. C.

Mr. Franklin Crowell, 512 E. Bank St., Salisbury, N. C.

Miss Sarah Culbreth, Dept. of Zoology, Duke Univ., Durham, N. C.

Mr. Joel E. Derrick, Spencer, N. C.

Mr. Richard A. Faust, 406 Mitchell Ave., Salisbury, N. C.

Mr. James Ferger, Dept. of Botany, N. C. State College, Raleigh, N. C.

Mr. Charles H. Flory, Box 1724, High Point, N. C.

Miss Frances K. Foust, U. N. C., Chapel Hill, N. C.

Mr. Kenneth H. Garren, Dept. of Botany, Duke Univ., Durham, N. C.

Mr. F. P. Gaskins, Phipps & Bird, Inc., 915 E. Cary Street, Richmond, Va.

Dr. John J. Gergen, Box 4771, Duke University, Durham, N. C.

Mr. Leland J. Gier, Campbell College, Buies Creek, N. C.

Mr. William Lanier Hunt, Box 169, Chapel Hill, N. C.

Dr. Thomas Kerr, Box 5035, N. C. State College, Raleigh, N. C.

Mr. F. J. le Clair, U. S. Soil Conservation Service, Chapel Hill, N. C.

Miss Jean Leitner, Dept. of Botany, U. N. C., Chapel Hill, N. C.

Dr. F. H. McCutcheon, Dept. of Zoology, N. C. State College, Raleigh, N. C.

Mr. Berlen C. Moneymaker, Box 134, Murphy, N. C.
 Mr. Grover Murray, Jr., Dept. of Geology, U. N. C., Chapel Hill, N. C.
 Dr. John M. Parker, III, Box 5171, N. C. State College, Raleigh, N. C.
 Mr. Avery Patton, Jr., Davidson College, Davidson, N. C.
 Mr. John H. Pierczynski, Bureau of Identification, Police Dept., Charlotte, N. C.
 Miss Caroline E. Powell, Boyden High School, Salisbury, N. C.
 Mr. F. P. Pratt, Jr., 528 Mitchell Ave., Salisbury, N. C.
 Mr. Donald D. Ritchie, Dept. of Botany, U. N. C., Chapel Hill, N. C.
 Miss Laurie M. Stewart, Dept. of Botany, U. N. C., Chapel Hill, N. C.
 Mr. H. W. Straley, III, Dept. of Geology, U. N. C., Chapel Hill, N. C.
 Dr. Herbert C. Tidwell, Wake Forest Medical School, Wake Forest, N. C.
 Mr. Henry Wagner, Catawba College, Salisbury, N. C.
 Mr. Woodrow Wilson, Dept. of Zoology, Duke University, Durham, N. C.
 Mr. W. F. Whitt, 521 Maupin Avenue, Salisbury, N. C.

"The following were reinstated to membership:

Dr. Lewis E. Anderson, Dept. of Botany, Duke Univ., Durham, N. C.
 Mr. Clifford Beck, Boyden High School, Salisbury, N. C.
 Dr. Sherwood Githens, Jr., Wake Forest College, Wake Forest, N. C.
 Mr. M. A. Hill, U. N. C., Chapel Hill, N. C.
 Dr. Z. P. Metcalf, N. C. State College, Raleigh, N. C.
 Dr. Arthur E. Ruark, U. N. C., Chapel Hill, N. C.
 Mr. S. O. Trentham, Mars Hill, N. C.

"The committee also reported the following losses during the year:

Lost by death: Dr. David H. Howard, Jr.
 Lost by resignation: Mrs. Margaret Stoughton Abell
 Dropped from the roll because of non-payment of dues: forty former members.

The Treasurer's report was as follows:

Financial Statement of the N. C. A. S. May 6, 1937

Receipts

Balance on hand April 23, 1936:

| | | |
|-----------------------|----------|----------|
| Savings Account..... | \$437.94 | |
| Checking Account..... | 132.06 | |
| Cash on hand..... | 4.00 | |
| Total balance..... | \$574.00 | \$574.00 |

Dues:

| | |
|-----------|--------|
| 1936..... | 164.00 |
| 1937..... | 218.00 |

Initiation fees:

| | |
|-----------|-------|
| 1936..... | 26.00 |
| 1937..... | 50.00 |

\$458.00 \$458.00

Replacement of returned checks..... 4.00

Interest on savings..... 10.71

Total receipts (Includes \$50.00 transferred from savings
to checking account)..... \$1,046.71 \$1,046.71

Disbursements

Stationery and printing..... \$62.02

Programs, 1937..... 28.50

Postage..... 2.36

Returned checks due to insufficient funds..... 4.00

Refund dues..... 2.00

Books for H. S. Essay Prize..... 16.34

Expenses for delivering prize..... 6.50

Journal of E. M. S. S., 1936 (\$250.00 from check-
ing and \$50.00 from savings)..... 300.00

Charges on bank balance..... 1.00

Clerical assistance..... 59.00

Sec.-Treas. commission..... 45.00

Refund Sec. dues..... 2.00

Telegrams..... .64

Telephone calls..... .40

Receipt books..... 1.03

Letter file..... .67

Stencils..... 1.87

Interval timer..... 5.32

Total disbursements..... \$538.65 \$538.65

Total balance May 6, 1937..... \$508.06

Savings Account

April 23, 1936..... \$437.94

Interest..... 10.71

\$448.65

Withdrawn Dec. 8, 1936 (\$50 transferred to check-
ing; \$50 cash to Journal)..... \$100.00

Balance May 6, 1937..... \$348.65 \$348.65

\$159.41

Dues on hand..... 8.00

Balance in checking account May 6, 1937..... \$151.41

The above report was made as of May 6, 1937.

Submitted by H. L. Blomquist, Secretary-Treasurer.

Audited May 7, 1937 by

J. B. Derieux,

G. Howard Satterfield,

A. F. Thiel.

"The committee accepted the invitation of the Faculty of the North Carolina State College of the University of North Carolina to hold the thirty-seventh annual meeting of the Academy in Raleigh.

"The executive committee made the following recommendations to the Academy:

1. That all bills presented in the treasurer's report be authorized and paid and that the report be printed when audited.

2. That Bert Cunningham be appointed to select the books to be presented to the winner of the High School Science Essay Prize, and that he be authorized to draw upon the treasury as much as \$25.00 for these books; and that the secretary be authorized to appoint a representative of the Academy to award the prize and draw upon the treasury for the payment of his expenses.

3. That hereafter the total expenditure for the High School Science Essay Contest be limited to \$25.00.

4. That the Secretary-Treasurer be authorized to pay the Elisha Mitchell Scientific Society \$300.00 for the publication of the 1937 Proceedings.

5. That C. W. Edwards of the Department of Physics, Duke University, and A. S. Wheeler, formerly acting chairman of the Department of Chemistry, the University of North Carolina, be made life members. Professor Edwards joined the Academy in 1902, was its president in 1903, and has been an enthusiastic and active member up to the present time. Dr. Wheeler joined the Academy in 1903, was its president in 1916, and has been an active member until last year when, because of ill health, he was compelled to retire from active duties."

The report and the recommendations of the executive committee were accepted and approved.

The auditing committee reported that the treasurer's report had been examined and was found to be correct.

The reports of the treasurer and auditing committee were accepted.

The committee on the Phipps and Bird Prize, consisting of Wm. F. Prouty, chairman, J. B. Derieux, J. P. Givler, Anne Pegram, O. J. Thies, and F. A. Wolf, reported as follows:

"The gold medal for the most noteworthy paper presented at the 1936 meeting was awarded to Dr. F. G. Hall, Duke University, for his paper, entitled 'Physiological Studies at High Altitudes.' The paper was awarded second place in the interstate academy contest."

The committee on high school science, consisting of Bert Cunningham, chairman, H. B. Arbuckle, Mrs. B. W. Wells, C. E. Preston, R. J. Campbell, and Mary Conrad Cleaver, reported as follows:

"Your Committee reports that it has carried on its usual activities during the year, including the Essay Contest.

There were 21 essays from 15 schools entered in the contest. The Judges, Dr. H. J. Oosting, Mrs. B. W. Wells, and Dr. C. E. Preston selected Miss Lucy Nelms of the Nashville High School as the winner of the prize. As in years passed this essay will be submitted to the High School Journal for publication. The Committee recommends that it be empowered to select the books for the prize (not to exceed \$20.00) and that the Secretary of the Academy appoint a representative of the Academy to personally present the award at the Commencement Exercises of the Nashville High School.

The Committee has carefully considered the continuation of this prize, and has reached the following decisions:

(1) The prize should be continued. The number of essays submitted in any one year is no evidence of the extent of the influence of the contest. New schools are constantly submitting essays, and most schools do not enter the contest in consecutive years. Many essays are prepared which never come to the Academy since each school is limited to three entrants. We recommend that the prize be awarded next year in the fields of Chemistry and Physics.

(2) The Committee, however, feels that the present procedure of widely circularizing the schools does not bring commensurate returns. It suggests that the procedure for next year be altered by substituting personal correspondence with comparatively few teachers for the present system of broadcasting. This could probably be done at no expense to the Academy other than the prize itself, which the Committee feels should not be lessened. We recommend that this procedure be followed.

The Committee has also considered the possibility of creating an interest in the Academy on the part of the high school teachers. It suggests that this might be partly accomplished by having a special section with its own program on Saturday morning for this group. It also thinks it might be possible to create greater interest on the part of

teachers if the sponsor of the winner of the Essay Contest were given some recognition such as honorary membership in the Academy for a year. The Committee, however, is not ready to make specific recommendations on either of these latter matters."

The report of the high school science committee was accepted and its recommendations approved.

The legislative committee, consisting of Z. P. Metcalf, chairman, W. L. Poteat, and Wm. F. Prouty, had no report at this time.

The elective appraisal committee, consisting of P. M. Ginnings, chairman, E. H. Hall, R. E. Coker, Mary Conrad Cleaver, C. W. Edwards, Karl Fussler, E. G. Purdom, R. N. Wilson, J. T. Dobbins, reported that the committee had had no requests for appraisal and, therefore, had no report to make.

The conservation committee, consisting of Charles E. Raynal, chairman, B. W. Wells, J. P. Givler, C. F. Korstian, W. C. Coker, and J. S. Holmes, made the following report which was prepared and read by J. S. Holmes at the request of the chairman of the committee:

REPORT OF THE CONSERVATION COMMITTEE OF THE NORTH CAROLINA ACADEMY OF SCIENCE

BY DR. J. S. HOLMES

The idea of conservation has undergone very considerable change since it was first linked with the better management of our natural resources some thirty years ago.

At the American Forestry Congress held in Washington, January 1905*, called by President Theodore Roosevelt "to establish a broader understanding of the forest in its relation to the great industries depending upon it; to advance the conservative use of forest resources for both the present and future need of these industries; to stimulate and unite all efforts to perpetuate the forest as a permanent resource of the nation," the word "conservation" does not seem to have been uttered, though foresters and lumbermen alike very fully discussed "conservative lumbering." By 1908, however, popular interest in forest conservation had become so keen that the same President "called together in conference the governors of all the States with their advisers, together with the presidents of the various national societies interested, and others, to discuss the broad question of the conservation of natural

* See Proceedings.

resources. As a consequence national and state conservation commissions were formed in all parts of the Union, and a new era of active interest in economic development seemed to have arrived,"* by the following year, 1909. At that time conservation was defined by Gifford Pinchot, the Chief Forester, as the wise use of our forests, waters, soils, and ores (mineral resources). A little later *wild life* was added as a fifth resource and subsequently the *human* resource was taken under the expanding wing of conservation.

With the experience of three decades we might now define conservation as the management of all our natural resources so that the greatest good to the greatest number over a long-time program will result. Since the great majority of the American people know little and care less about *any long-time* program it will be many more decades before real conservation can be put into general practice. However, if organizations such as this Academy understand the need and advocate the practice of conservation, the future leaders of State and Nation will be better informed, and inspired to do the right thing by the abundant gifts with which this country has been blest.

We now seem to have two schools of conservationists, each of the opinion that its point of view is the true one. The practical man bases his opinions on the *economic* use of our natural resources; and *sometimes* this use is based on *commercial demand* rather than on *public need*. And though this man, who favors "wise use," at times fails to look far enough ahead, his policy, improved with knowledge, must no doubt govern the management of the bulk of our resources.

Strongly opposed to this point of view is the aesthetic conservationist, he who would preserve the beauty of the forest or waterfall by prohibiting or greatly limiting its economic use. This man puts recreation and enjoyment first and on him we depend chiefly to save our beauty spots; while the other puts ahead the plain drab need of making a living. Neither questions the *need* of the other but each insists that *his* policy is the more important.

If this question is to be determined by the pressing immediate needs of the *private landowner* and the *two-year* view of succeeding legislatures, we shall, I fear, continue to neglect the *long-time* view and our conservation practice will continue to remain ineffective until the resources have disappeared.

In addition to these two opposing view points of conservation there is to my mind a third which may be considered a combination or a com-

* Fernow's History of Forestry, page 431.

promise or a way out, which we might call the *ethical* point of view. Many people who have thought as well as searched, have faith and conviction that there is a master mind behind this matter and that there is a divine plan of growth, improvement, and development which we call evolution. Recognizing the existence of such a plan, should we not work with Nature as closely as our actual needs will allow, protecting and preserving for future generations all economic and aesthetic resources in at least as good condition as they came to us from the past? By destroying or even materially reducing in one generation any such natural resource we are defrauding our descendants to infinity, which is worse than simply defrauding our neighbour.

However, let us now come down to a few present day problems. First a few words about the policy of Stand Improvement on National Forests, which was the primary cause of the appointment of this committee two years ago. The National *Forests* are administered on an *economic* basis as opposed to the *aesthetic* interests of the National *Parks*. The primary values of the National Forests are regulation of stream flow and production of timber. The large majority of forest land, both publicly and privately owned, *must* be handled as a *crop* for the production of revenue, and as the farmer considers those plants weeds which injure the desired farm crop, so the forester considers those species weeds which interfere with the development of the *salable timber* crop. This is the justification for the cutting out of dogwood and other of the less valuable species in order to favor the more valuable ones, which is, as just mentioned, an accepted practice by farmers growing annual crops. This *policy* must meet our approval, though sometimes its *practice* does not. Forestry in this country is in its infancy and no owners of forest land are in a position to act with full knowledge of what the final results will be, as full information on growth, future markets, and other factors is not yet available.

The conservation of our scenery is a problem that our people are only just waking up to. The efforts of the various states to secure tourist travel and desirable settlers emphasize our need for the protection of what natural scenery we have. While economic necessity seems to demand the construction of highways, telephone lines, and other improvements across the landscape, a growing desire for beauty will demand the restriction of these and other so-called improvements in many areas. It is only through the education of the public that improvement in this respect will come about and the primary object of this report is to ask the coöperation of members who are educators in emphasizing the principle of conservation in every line of endeavor.

The need for conserving the supply of our food fishes is well illustrated by the very serious decline in our shad fisheries. We have expended large sums of public as well as private money in the construction of locks, dams, etc., across larger streams without any thought or care that this would keep the shad from their spawning ground. Unless every precaution is taken similar destruction of other food fishes will be caused in our waters by the rapidly expanding pulp industry and the consequent pollution of our streams.

Another serious problem is the destruction of wild life under ill-advised State and Federal laws favoring the group known as game. Conservation to many sportsmen, as well as to the makers and purveyors of arms and ammunition, seems to mean the artificial production of a few game birds and the destruction of all birds and animals which they consider inimical to the sportsman's interest and class as predators and vermin. A recent very determined attempt to secure legislative authority in one of our eastern counties for the indiscriminate hunting and killing of bear, muskrats, rabbits, and foxes at any time and without a license, and to allow the poisoning of crows and jackdaws is a nearby instance of misguided enthusiasm for the conservation of one resource at the expense of another.

Wild life administrators are rapidly coming to the conclusion that the artificial production of game birds is no solution to the growing scarcity of game species. The conservation of their natural feeding and breeding areas, with the restoration, so far as possible, of what we refer to as the "balance of nature" is the only practicable policy.

There are many problems confronting public officials responsible for the administration of our natural resources which must depend on scientific research for their solution. North Carolina will continue to count on the cooperation of the scientists of the State in such public service. However, the great task before us is the conversion of a wasteful and indifferent public, to one recognizing its duties to the future, before it is too late.

Conservation cannot be superimposed on a State or Nation by just changing the name of one Federal Department, which has been notorious for its dissipation of our natural resources, to the Department of Conservation, and taking to itself other government agencies already deeply interested in and practising various phases of conservation; but the principle must be drilled or instilled into our people, especially our young folks, as they gradually accumulate knowledge about our re-

sources. While every citizen has his responsibility for handling those resources which come into his care in a conservative way, realizing his full responsibility to the future, it is more especially the task of those who are studying nature and are teaching the youth of our land to emphasize at every opportunity the responsibility which the handling of our natural resources places upon each one of us. For this reason I feel that this Academy can well encourage among its members not only research and study of our natural resources but coupled with it the training of their students and the public in the *wise use* of these resources."

The report of the conservation committee was approved and the secretary was authorized to order 250 reprints of this report when published.

The committee on the A. A. A. S. grant, consisting of R. E. Coker, chairman, M. L. Braun, O. J. Thies, J. G. Boomhour, and Eva G. Campbell, reported:

"That the A. A. A. S. grant for 1936 was awarded to Mr. D. S. Correll for his 'Studies on the orchids of the southeastern states,' and that the grant for 1937 be awarded to L. G. Willis for the continuation of his studies on 'The oxidation-reduction equilibrium in soils.' A special request was made that more applications be sent in for this grant."

The report of the committee on the A. A. A. S. grant was accepted.

A memorial report honoring the late Dr. David H. Howard, Jr., was presented:

DR. DAVID HALBERT HOWARD, JR.

"David Halbert Howard, Jr., passed to his eternal home on April 26, 1936. In his passing his devoted family in Virginia was overwhelmed in grief, Davidson College suffered an irreparable loss, and this Academy has lost one of the most brilliant young men who ever came to North Carolina to contribute his energies to the advancement of science.

"David Howard was born in Lynchburg, Virginia, on June 23, 1906, with a social background that fitted him for rare attainment in scholastic achievements.

"At the age of eighteen years he graduated from the Lynchburg High School at the head of his class.

"The following fall he entered Davidson College, became a popular

student on the campus, and early in his college course was outstanding in literary and scholastic activities.

"He was a member of the Eumenean Literary Society, the International Relations Club, Omega Phi Alpha, Gamma Sigma Epsilon, and Phi Beta Kappa. He was a leader of the Sigma Phi Epsilon Social Fraternity and contributed largely toward placing this Fraternity first in scholarship at Davidson College.

"He graduated at Davidson as valedictorian in his class and won many honors during his college course, notably membership in Phi Beta Kappa and likewise in Gamma Sigma Epsilon, an honor Chemical Fraternity. His leadership in the above Chemical Fraternity brought to him the high honor of being elected as a Grand Officer.

"After his graduation in college he accepted a place as instructor in Chemistry at his alma mater which gave him opportunity of making this one of the most fruitful years in his study of Chemistry. When he completed this year's work, he took up graduate work at Cornell University. He was a teaching assistant at Cornell in 1930 and received a fellowship each year afterwards until he was awarded his Ph.D. degree in 1933.

"Conspicuous honors fell to him at Cornell, being elected to membership in Sigma Xi, Alpha Chi Sigma, and Phi Kappa Phi.

"From 1933 to 1935 he was an assistant professor at George Washington University and received a call as associate professor of chemistry at Davidson College in 1935.

"His rare combination of geniality, sincerity, high ideals as a teacher, and his unselfish zeal in helping and encouraging students made him a much liked teacher from the very start, and he promised to make a professor in the college, outstanding in character, service, and personality.

"As proof of his zeal in science, we cite the fact that during his first year as professor at Davidson he joined the North Carolina Academy of Science and the American Chemical Society.

"Already he, in conjunction with one of his colleagues at George Washington University, published a laboratory manual in general chemistry and a number of articles were published in the Journal of the American Chemical Society.

"Truly a gracious and beautiful character has passed on to his award.

"May his unselfish conduct, his uplifting influence, and his noble example continue to bless his loved ones and his friends through the eternal years.

"The Academy deeply mourns his loss and extends sincerest sympathies and condolence to his bereaved family."

H. B. ARBUCKLE,
W. L. PORTER,
O. J. THIES, JR.
Committee.

Bert Cunningham, representative of the Academy to the A. A. A. S. reported as follows:

"Some thirty representatives of the various academies attended the Academy Conference for 1936-37. The program consisted largely of the reports of the activities of the various academies in addition to their usual annual programs. A number of academies reported especially helpful relations with Junior Academies. It became evident during the progress of the meeting that there were important problems which academies should face, such, for example, as securing funds for research, initiating suitable legislation for academic freedom, and providing proper publication for scientific work. The dinner given by the A. A. A. S. was well attended. Dr. Bilsig was re-elected as secretary for a three year period.

"The Council this year handled only matters of routine. The organization of a southern-eastern section of the Association was not brought up. A full report of the Council actions has already been printed in *Science*. Richmond has been selected as a meeting place for 1938-39 and it is to be hoped that the members of this Academy will aid in every way possible to make that meeting a success."

The general resolutions committee, consisting of J. P. Givler, chairman, R. J. Campbell, and E. E. Randolph, presented the following resolution:

"The North Carolina Academy of Science wishes to express its great appreciation of the manner in which it has been entertained by Catawba College.

"We desire to express our thanks for the hospitality of the College at the complimentary supper, at the dramatic entertainment, and the reception, and for the gracious manner in which the students of Catawba College assisted in the registration and in the posting of the progress of the papers of the different sections during the meeting."

The nominating committee submitted the following nominations:

President: W. E. Speas, Wake Forest College;

Vice-President: M. L. Braun, Catawba College;

New member of the Executive Committee (for three years): R. F. Poole, North Carolina State College of the University of North Carolina;

Two new members of the Committee of the A. A. A. S. Grant (for three years): G. W. Hargitt, Duke University, R. N. Isbell, Wake Forest College;

Representative to the A. A. A. S.: Bert Cunningham, Duke University.

The nominations were accepted and the secretary was instructed to cast the ballot of the Academy for the nominees.

The president then announced the appointment of the following committees:

The Legislative Committee: H. F. Prytherch, chairman, B. W. Wells, W. F. Prouty.

The Conservation Committee: J. S. Holmes, chairman, J. P. Givler, C. F. Korstian, W. C. Coker, H. J. Oosting.

The High School Science Committee: Bert Cunningham, chairman, H. B. Arbuckle, Mrs. B. W. Wells, C. E. Preston, R. J. Campbell, Mary Conrad Cleaver.

The Committee on the Phipps and Bird prize: J. N. Couch, J. B. Derieux, J. P. Givler, Annie Pegram, O. J. Thies, F. A. Wolf.

The Elective Appraisal Committee: to stand with no changes.

The business meeting then adjourned.

At 6:30 the Academy and visitors were entertained at a complimentary barbecue supper on the College Campus.

At 7:30 the local dramatic club entertained the Academy with a one act comedy, "There are none of them perfect."

At 8:30 the evening meeting was held with the vice-president, C. F. Korstian, presiding. The address of welcome was made by Howard R. Omwake, president of Catawba College. This was followed by the presidential address, entitled "The Interdependence of the Sciences," by the retiring president P. M. Ginnings.

On Saturday morning the Academy met in sections. Vice-president C. F. Korstian presided over the General Section; J. M. Clarkson over the Mathematics Section; and Milton L. Braun over the Physics Section. The North Carolina Section of the American Chemical Society did not hold a meeting at this time and the election of officers was done by mail.

The following officers were elected by the respective sections:

Mathematics Section: Chairman, J. J. Gergen, Duke University; Secretary, Archibald Henderson, The University of North Carolina.

Physics Section: Chairman, J. B. Derieux, North Carolina State College; Secretary, F. W. Lancaster, North Carolina State College.

North Carolina Section of the American Chemical Society: Chairman, W. C. Vosburgh, Duke University; Vice-President, R. N. Isbell, Wake Forest College; Secretary-treasurer, E. C. Markham, The University of North Carolina; Councilors, D. G. Hill, Duke University; R. W. Bost, The University of North Carolina; Executive Committee, the officers and C. S. Black, Wake Forest College; W. A. Reid, North Carolina State College; Edward Mack, the University of North Carolina; W. E. Jordan, North Carolina State College.

The following papers were presented. Those marked with * appear in full in this issue; those marked x are abstracted with the Proceedings; those marked † were read by title.

GENERAL SECTION

Additional Sources of Androsterone. M. D. KRITZER and BERT CUNNINGHAM, Duke.

†x *Caves of Yucatan.* A. S. PEARSE, Duke.

†x *A Photoelectric Device for Rapid Measurement of Leaf Areas* (Lantern). PAUL J. KRAMER, Duke.

**Development of the Perithecium of the Ascomycete, Sordaria fimicola* (Opaque Projector). DON RITCHIE, U. N. C.

Paraffin Crystals: Spiral and Other Forms (Lantern). CHARLES M. HECK, N. C. State.

Methods of Research in Soil Fertility (Lantern). L. G. WILLIS, N. C. State.

**A Fungus that Catches Nematodes* (Lantern and Opaque Projector). JOHN N. COUCH, U. N. C.

A New Species of Rozella Parasitic on Allomyces (Lantern). FRANCES K. FOUST, U. N. C.

Published in full in this Journal **53**: 197-204. 1937.

Surface Tension of Water at Low Pressures (Lantern). A. A. DIXON, N. C. State.

The Primary Wall of the Cotton Fiber (Lantern). DONALD B. ANDERSON, N. C. State.

Secondary Wall Deposition in the Cotton Hair (Lantern). THOMAS KERR, N. C. State.

Survey of the Fungi of the Duke Forest. FREDERICK A. WOLF, Duke.

Cytological Observations on Thraustotheca clavata (Lantern and Opaque Projector). LELAND SHANOR, U. N. C.

Published in full in this Journal **53**: 119-136. 1937.

x *Direct Appraisal of Available Cellulose in Wood for Pulp Yields.* E. E. RANDOLPH, N. C. State.

- x *The Wind as a Sorting Agent*. G. R. MCCARTHY and J. W. HUDDLE, U. N. C.
- x *Endosperm Development in the Araceae* (Lantern). MURRAY F. BUELL, N. C. State.
- Varietal Differences in the Vitamin C Content of Tomatoes* (Lantern). G. HOWARD SATTERFIELD and FRANCIS TRIPP, N. C. State.
- Oedocladium Lewisii, a New Green Alga* (Lantern). L. A. WHITFORD, N. C. State.
- Mileage of Different Gasolines and at Different Speeds* (Opaque Projector). J. B. DERIEUX, N. C. State.
- Wintering of Honeybees in North Carolina: Some Recent Results* (Lantern). F. B. MEACHAM, N. C. State.
- x *Influence of Parathyroid Extracts on Growing Bone*. ROBERT B. BURROWS, Elon College.
- x "Permian." WILLARD BERRY, Duke.
- x *Notes on Buckleya and Pyricularia (Buffalo-Nut)*. H. R. TOTTEN, U. N. C.
- Archaeology in North Carolina*. JOFFRE L. COE, U. N. C.
- x *Granite Intrusion in Eastern Wake County, North Carolina*. JOHN M. PARKER, N. C. State.
- x *Volcanic Rocks in the Eastern Piedmont of North Carolina*. J. L. STUCKEY, N. C. State.
- x *Control of the Oyster Drill (Motion Picture)*. HERBERT F. PRYTHERCH, U. S. Bureau of Fisheries, Beaufort.
- Effect of Some Fats on Gastric Motility* (Lantern). HERBERT C. TIDWELL, Wake Forest.
- x *Asheville, a Study in Urban Geography*. MARTHA E. NORBURN, Baltimore.
- x *A New Qualitative Test for Selenium*. H. A. LJUNG, Guilford College.
- Air Cells in Snow Crystals and Other Forms of Ice* (Lantern). CHARLES M. HECK, N. C. State.
- x *Lactogenic Substance in Reptile Pituitaries* (Lantern). BERT CUNNINGHAM, Duke.
- Utilization of Whole Cotton*. FRANK K. CAMERON, U. N. C.
- x *The Nomenclature of Fractures*. GROVER MURRAY, U. N. C.

MATHEMATICS SECTION

- The Symbolic Calculus of Two Variables Applied to the Heat Equation*. F. G. DRESSSEL, Duke.
- x *On the Classification of Collineations*. J. W. LASLEY, JR., U. N. C.

Lebesgue Integrals and Riemann Sums. J. J. GERGEN, Duke.

**A Century Old Problem in Euclidian Geometry: A Basic Study with Ten New Solutions.* ARCHIBALD HENDERSON, U. N. C.

x *Sets of Conjugate Matrices.* E. T. BROWNE, U. N. C.

PHYSICS SECTION

x *Chemical Analysis by Means of Infrared Spectra* (Lantern). E. K. PLYLER, U. N. C.

The Effect of Pressure Changes on the Appearance of the High Frequency Glow Discharge (Lantern). SHERWOOD GITHENS, JR., Wake Forest.
Gasoline Mileage in Terms of Octane Rating and Volatility Number (Opaque Lantern). J. B. DERIEUX, N. C. State.

x *Cosmic Ray Showers Produced in Large Thickness of Various Materials* (Lantern). KARL Z. MORGAN and W. M. NIELSEN, Lenoir-Rhyne and Duke.

x *Plastic Give and Recovery in Stretched Neoprene* (Lantern). MILTON L. BRAUN, Catawba.

EXHIBITS

Photomicrographs of Paraffin Crystal Forms. CHARLES M. HECK, N. C. State.

Destruction of Oysters by Drills. HERBERT F. PRYTHERCH, Beaufort.

Influence of parathyroid extracts on growing bone. ROBERT B. BURROWS, Elon College.

North Carolina Shore Birds. FRAZER G. POOLE, Catawba College.

The following abstracts have been received:

Caves of Yucatan. A. S. PEARSE.

The caves in Yucatan are of particular interest because they have for centuries served as sources of water for Mayas and therefore played an important rôle in the development of Mayan culture. During the summer of 1936 twenty-seven caves were studied. Air temperatures ranged from 22.0° to 27.9°C°; water temperatures, 22.0° to 27.2°C°; humidities, 90.2 to 100%. Five species of fungi have been cultured by Dr. F. A. Wolf from debris brought from caves. Nineteen papers have been prepared by specialists on the animals collected, which include land planarians, trematodes, cestodes, nematodes, oligochaetes, leeches, gastropods, copepods, ostracods, shrimps, isopods, schizopods, thysanurans, collembolans, cave crickets, cockroaches, beetles, flies, mosqui-

toes, ants, wasps, parasitic hymenopterans, fleas, millipedes, centipedes, spiders, mites, ticks, whip scorpions, pseudo-scorpions, fishes, lizards, snakes, and mammals. Most of the types collected were troglonexes, but there were some troglaphiles and troglóbites. Blind species included shrimps, isopods, schizopods, collembolans, ants, millipedes, mites, brotulid fishes, and a symbranchid eel.

A Photoelectric Device for Rapid Measurement of Leaf Areas. PAUL J. KRAMER.

By the use of a photoelectric apparatus leaf areas can be measured much more rapidly and with nearly as great accuracy as by any of the methods ordinarily used. The leaves are placed on a ground glass plate interposed between a light source and a photoelectric cell which is connected to a microammeter. The deflection of the microammeter is inversely proportional to the amount of light intercepted and hence to the area of the leaf or leaves. By plotting the microammeter readings for pieces of leaf of known area a curve can be drawn from which the area corresponding to any observed reading can be quickly determined. The apparatus described by the writer has two improvements not found in those previously described. A cone of polished aluminum was inserted between the ground glass plate and the photoelectric cell. This concentrates the light on the cell so that a light source of much lower intensity can be used. It also reduces positional error. A resistance was inserted in the light source circuit permitting adjustment of the light intensity to compensate for variations in line voltage. These features have appreciably increased the accuracy and made it possible to use the apparatus in spite of considerable variations in intensity of the light source.

Direct Appraisal of Available Cellulose in Wood for Pulp Yields. E. E. RANDOLPH.

The interest in pulp and papermaking in the South has been greatly increased during the past year. At present companies are erecting ten large new pulp mills; one in Virginia, one in North Carolina, two in South Carolina, two in Georgia, two in Florida, one in Arkansas and one in Texas. Two of these mills are just beginning operation. It is reported that the total cost of these mills is over sixty-six million dollars. Negotiations are in progress concerning the building of other mills for pulp and news-print paper in the South. There are several reasons for such large investments in new paper industries. The longer growing

season and the fertile soil account for the rapid growth of timber to marketable size within about twenty years, whereas in northern latitudes trees must become thirty-five or forty years of age before they are of marketable size. The climate assures all year round operation at full capacity. There are ready markets. There is a plentiful supply of suitable water. There are good highway, railway, and water shipping facilities. There is a sufficient supply of skilled and common labor. Already an enormous supply of pulp and paper is being produced in the South. In North Carolina, for example, is located one of the largest paper mills in the world producing daily between four and five hundred tons of pulp and paper and there are in active operation also two other paper mills in the state.

The essential part of wood for pulp making is cellulose. This material forms cell walls of plants. There are in plants and trees various other constituents besides cellulose. In order to make good paper, it is necessary to remove the other constituents from the wood and leave the cellulose in as pure form as possible. In the mechanical process for making pulp none of the other constituents is removed from the cellulose. The total weight therefore of the dry log goes into pulp. In the chemical process, however, the other constituents are removed from the cellulose to a greater or less extent so that from 50 to 60 per cent of the weight of the wood yields pulp.

The purchasing agent of the company buys lots of wood on the basis of the cellulose content in the wood which can be realized in the digester yields. It is important that a reliable method may be available to determine the cellulose content of the wood which may reasonably be expected to represent the yields which can actually be made on the plant runs in the mill. Different methods have been devised for determining cellulose content of the plants. The most important of these methods is the Cross and Bevan method. Unfortunately, this method takes considerable time and does not always gives yields which can be duplicated in the plants. The importance of a method which does give results comparable to what may be expected in plant yields is apparent. The object in this investigation was to see if a direct method could be devised which would be accurate, dependable, and capable of being performed in a short time.

This research was carried on in the Chemical Engineering Department of State College during the year and satisfactory results were obtained. The plan pursued is to build a small constant temperature oil bath regulated by automatic control so that the temperature does not vary

more than one-half of one degree. Small stainless steel digesters are used. The wood under examination is sawed with a coarse circular saw and the sawdust is carefully screened. It is then placed in autoclave with the digestive liquor. In this work the sulphite liquor was used. Time is gained by using sawdust rather than the commercial size chips in the digester. The digestive liquors penetrate the chips in a digester by capillary action. It is slow in permeating the interior chips. By having sawdust instead of chips the penetration of the liquor is rapid. In this way an actual cook of the wood can be made within three or four hours whereas the Cross and Bevan method required two to three days work. The outfit is reasonably small and portable and can be taken from place to place and used wherever electric current is available to run the motor and operate the automatic electric heat device. The purchaser therefore can with a small amount of trouble make a determination on a cellulose content of the wood which will give him such information that he can pay the farmer a price on the assurances that he can duplicate the yields in cellulose in the plant operation. The farmer can be assured that he is receiving the full pay for the value of his wood. A sufficient number of digestions and examinations were made to satisfy ourselves that this direct appraisal method gives accurate and dependable results.

The Wind as a Sorting Agent. G. R. MCCARTHY and J. W. HUDDLE.

It is well known that the grains of dune sands are rounder than the grains of beach sand, and this difference is usually ascribed to greater abrasion (caused by mutual attrition) suffered by the wind-blown sands. Field evidence shows that dune sands which have been shifted too short a distance by the wind to have suffered appreciable abrasion are nevertheless distinctly rounder than the beach sands from which they were derived. Experimental evidence has now been obtained which shows that the superior roundness of dune sands is largely due to a selective action by the wind, the rounder grains moving more readily and hence being concentrated in the dunes. This movement under the influence of the wind does not seem to be one of rolling, but rather one of jumping, or saltation, the individual leaps of the rounder grains carrying them farther than the more angular ones.

Endosperm Development in the Araceae. MURRAY F. BUELL.

Of the three types of endosperm development, the nuclear type in

which endosperm formation is initiated by a period of free nuclear division, is the most common in the angiosperms. The cellular type, in which each division from the very first is accompanied by wall formation, is much less common. A third, the basal apparatus type, is often spoken of as the Helobiae type since it is so characteristic of that group. In this type at the first division of the primary endosperm nucleus a wall is laid down dividing the embryo sac into two cells. In each of the resulting two cells further endosperm development is different. The micropylar cell may have nuclear or cellular endosperm while the basal cell may remain undivided and even enlarge into a huge haustorial cell. Nuclear endosperm may be produced or even cellular endosperm but always this basal apparatus is of distinctly different nature from the rest of the endosperm. That the basal apparatus may sometimes be even more fundamentally different is indicated by a study of *Acorus Calamus* in which the basal cell is apparently cut off before the pollen tube enters. If double fertilization does occur then the tissue produced in the upper cell is triploid and that in the basal cell is diploid.

Influence of Parathyroid Extracts on Growing Bone. ROBERT B. BULLOWS.

Studies were made of the effects of two different parathyroid extracts on the bones of immature albino rats with the purpose of observing the detailed changes in the bones and the probable method of mobilization of the excess calcium found in the blood during hyperparathyroidism. Parathyroid extracts cause a condition of osteitis fibrosa just as does hypersecretion of the parathyroid glands. This, however, soon gives way to a different condition, called "marble bone disease," which consists of hypercalcification rather than a fibrous state, and is not characteristic of true hyperparathyroidism.

Although Collip's and Hanson's extracts are supposed to affect blood calcium to the same extent, they do not affect bone to the same degree, when given in equivalent doses. Collip's is much more pronounced in its effects.

Osteoclastic activity appears to follow decalcification and not to be the cause of it during the production of osteitis fibrosa. The underlying causes of decalcification, anti-extract formation, production of marble bone disease, and subsequent events appear to be chemical in nature and to be due to the impurities in the extracts. Since parathyroid extracts do not duplicate true hyperparathyroidism, but bring

into prominence several anti-body-like activities, they should be used with more caution in therapeutics and conclusions derived from experimental work should be evaluated carefully.

"Permian." WILLARD BERRY.

The Permian in eastern North America was separated from the Upper Carboniferous in the year 1880 by Fountain and I. C. White on the basis of the fossil plants found in the Cassville shale which lies on top of the Waynesville No. 11 coal. It has been recognized that the location of this boundary was more or less arbitrary but at that time no other evidence was available. Recently this boundary has been subject to discussion and various lines of evidence have been advanced either to prove its location or to change it, up or down in the section. In this instance the author advances the possibility that due to the appearance of red beds in the middle Conemaugh the boundary should be moved down to the base of that horizon; by explaining the occurrence of the red beds as evidence of organic movement or of a climatic change consequent upon organic movements of considerable magnitude to the eastward.

Notes on Buckleya and Pyrularia (Buffalo-nut). H. R. TOTTEN.

Herbarium specimens were shown and the history and distribution of what W. W. Ashe called "our largest known American parasite," *Buckleya distichophylla* (Nutt.) Torr., was reviewed. Previously it had been reported only from the banks of the French Broad River in east Tennessee and in North Carolina and from the banks of the Pigeon River in Haywood County, North Carolina. A new location and state for this interesting plant was reported, viz., the bank of Dismal Creek in Bland County, near the Giles County line, in Virginia, where with a class from the Mountain Lake Biological Station of the University of Virginia, we found it growing under *Tsuga canadensis*.

Herbarium specimens were also shown of *Pyrularia pubera* Michx. This plant has a rather wide distribution through the mountains and upper piedmont of our region. It is found associated with quite a variety of plants and its rather elaborate underground runner system, connecting what at first appears to be separate plants, shows little connection with the surrounding plants. While not questioning the proof that it is sometimes a parasite, the question is again raised as to whether it is an obligate parasite or can also exist independently.

Granite Intrusion in Eastern Wake County, N. C. JOHN M. PARKER, III.

Preliminary work on the detailed mapping and description of the geology of Wake County, by Dr. J. L. Stuckey and the writer, has brought to light a number of structural features related to a Carboniferous granite batholith. The eastern third of the county is underlaid by biotite granite, intruded into Pre-Cambrian gneiss, schist, slates, and metamorphosed volcanic rocks. The boundary lines of the granite are regular, straight, and nearly always parallel to the foliation in the walls. The country rock has been much altered by lit-par-lit injection between the laminations, by soaking with granitic solutions, by the intrusion of a net-work of pegmatite dikes and quartz veins, by the development of fracture cleavage dipping at 20-25° toward the intrusion, and by displacement along small thrust faults in the same position. The granite contains abundant inclusions in a zone some two miles wide along both sides. Individual inclusions range up to half a mile wide by two miles long, and are granitized and crumpled. The planes of flowage of the magma are well preserved in the parallel orientation of biotite flakes, tabular feldspar crystals, and small inclusions. They tend in most instances to be vertical or to dip eastward at high angles, and to be parallel to the contacts and the foliation of the walls. The intrusion is composite, being in places gray and even-grained, in others gray, coarse and porphyritic, and in one area pink from abundant microcline. The presence of zones of excellent flow banding and marginal thrusts in the interior of the granite indicate that these varieties represent separate intrusions in a composite batholith.

Volcanic Rocks in the Eastern Piedmont of North Carolina. J. L. STUCKEY.

Volcanic rocks, all of pre-Cambrian age and the same general physical characteristics, occur in three distinct areas along the eastern part of the Piedmont Plateau.

First, and most important, is the slate belt which crosses the state in a general northeast-southwest direction and varies in width from 20 to 60 miles. The slate belt includes parts or all of the counties of Union, Stanley, Montgomery, Davidson, Randolph, Chatham, Orange, Durham, and Person.

The second area is a narrow zone along the western parts of Wake County, crossing the east central part of Granville and the northwest corner of Vance.

The third area is represented by a series of out-crops along the western edge of the Coastal Plain. These out-crops are exposed chiefly along the river valleys eastward as far as Weldon, Rocky Mount, Goldsboro, and Erwin.

Recent field work by the writer indicates that the extent of these volcanic materials along the western part of the Coastal Plain is much more extensive than indicated on any published map or in any available reports. It seems very probable that the whole western part of the Coastal Plain as far east as Erwin, Goldsboro, Rocky Mount, and Weldon is underlaid with volcanic rocks. Between streams these are covered with a thin veneer of Coastal Plain material. The strike-and-dip of the cleavage in these rocks varies widely from place to place, indicating that the processes of metamorphism have been exceedingly complex in these areas.

Control of the Oyster Drill. H. F. PRYTHERCH.

Two species of marine snails, *Urosalpinx cinerea* and *Eupleura caudata* destroy seed and adult oysters valued at several million dollars annually. Investigations conducted in the Seaside region of Virginia and in North Carolina show that control of these drills or borers is possible by means of traps and by their removal during oyster culling operations. Drills are strongly attracted to seed oysters which are used as bait in traps made of chicken wire. The traps are arranged at intervals of 10 feet on a 250 foot rope line, which is fished weekly and the drills removed by vigorous shaking.

In experimental trapping operations a total of 320,880 drills (78.6 *Urosalpinx* and 21.4 *Eupleura*) were collected by the use of 4,750 traps over a period of 3 months. The most efficient procedure is to operate each trap line in successive radial positions with one end of the line stationary. By shifting the line 45 degrees after each fishing it is possible to remove virtually all of the drills from 4 acres of bottom in approximately 3 months.

Field studies of the drill population on representative tidal flat areas indicate that there are over 35,000,000 of these pests on the public and private oyster beds of the Seaside region. Studies of the rate of feeding of drills, conducted under natural conditions show that 100 drills can destroy over 125 seed oysters and 90 adult oysters in one month. Therefore, it is possible for the drills of this region to destroy nearly half a billion seed and adult oysters in one summer season. The depleted condition of the natural beds and the poor yield from oyster

cultivation operations in this section is due primarily to the destructive habits of these pests.

Asheville: A Study in Urban Geography. MARTHA E. NORBURN.

Although so isolated by mountain barriers that settlement was delayed until after the Revolution, Asheville has become a cosmopolitan city. It owes its growth to its commanding position at the convergence of communication routes in the heart of the Southern Appalachians.

English and Scotch-Irish pioneers penetrated rugged passes in the rim of mountains; followed animal and Indian trails whose pattern was determined by range, gap, and stream; built cabins; and, in 1792, organized Buncombe County.

The county seat, Morristown, was located near the confluence of the Swannanoa and French Broad rivers and at the crossing of the east-west and north-south trails. Population increased through the desire for land; through health seekers who found the climate of subdued mountains in the Virginia belt beneficial; and through scientists who came to study landforms and vegetation.

In 1797, the village of Morristown became the town of Asheville. Stock driving from the Mississippi Basin to the Atlantic Seaboard, the tobacco boom, and the railroad combined to increase population. In 1883, Asheville was created a city. Its growth has since been augmented by industries and by roads which serve the National Forests and the Great Smoky Mountain National Park.

The business zone has been altered to accommodate traffic, the "Square" dedicated to the crossing of highways, and a mountain tunneled. The pattern is still radial and conforms to the topography.

A seasonal population travels the roads. However, as the gaps and ancient trails control the communication pattern, the descendants of the English and Scotch-Irish pioneers determine the permanent pattern of population.

A New Qualitative Test for Selenium. H. A. LJUNG.

Within the past few years the element selenium has commanded considerable attention. It has been shown that plants take up selenium from soils in which it occurs, and that the plants which have taken up the selenium are toxic when used as foods for animals. Dr. Knight,¹ Chief of the United States Bureau of Chemistry and Soils, tells interestingly of the selenium problem in this respect. He relates that such

¹ Knight, H. G.; *Sigma Xi Quarterly* 25: No. 1, 1937.

diseases as "alkali disease" and "blind staggers" are due to selenium poisoning, and that thousands of cattle and sheep succumb annually from eating plants which contain selenium. Dr. Franke of the South Dakota Experiment Station, now deceased, also writes of his experiences and researches on this problem.²

In addition to the attention selenium has attracted from this quarter, it has found use in the production of stainless steels.

This great selenium problem of the west and the uses of selenium in steels has attracted the attention of the analytical chemist, and as a result new methods for the qualitative detection and quantitative determination of the element have been developed. The methods commonly employed are the distillation method using bromine-hydrobromic acid solution, and the reduction methods using such reducing agents as sulfur dioxide, potassium iodide, etc. The reducing agents commonly employed are unsuitable as standard solutions, hence a reagent which is stable and can be conveniently standardized would be more desirable.

The use of the thiocyanate ion as a standard reducing agent in other analytical reactions has suggested its possible use as a reagent for the reduction of selenious acid. No references are available in the literature concerning such a reaction, and no theoretical considerations are available concerning the possibilities of the reaction taking place, hence it was necessary to try out the possibility experimentally. It was found that the thiocyanate ion is capable of reducing the selenite ion in solutions acid with hydrochloric acid. (The reaction also takes place in solutions acid with sulfuric acid; however, as yet this work is incomplete. A report will be made later concerning the reaction in sulfuric acid solutions.) The selenium is precipitated as red metallic selenium in high concentrations of selenium; as a yellowish green precipitate in low concentrations; and as an almost white precipitate in exceedingly low concentrations. The precipitate is usually finely divided, remaining suspended for almost an hour. Hydrogen sulfide is also one of the end-products of the reaction. Unfortunately this work has not progressed far enough to permit the writing of an equation to represent the reaction between the ions. However, work has been done which reveals some of the properties of the reaction, as follows:

(1) The reaction takes place rather slowly and incompletely in solutions less than six normal in hydrochloric acid. At higher concentrations of acid the reaction proceeds rapidly and quantitatively.

(2) The reaction takes place only slowly at room temperature—re-

² Franke and Painter, *Ind. Eng. Chem.* **29**: 591-95, 1937.

quiring more than thirty hours for completion when as little as 2.5 mg. of selenium is present. The reaction proceeds rapidly at temperatures above 60°C.; and is quite rapid at the boiling-point of the solution—requiring only 20–30 seconds for completion with small quantities of selenium.

(3) Volumetric relationships indicate that there is a stoichiometric ratio between the ions involved.

(4) The reaction may be carried out in the presence of the common metals, with the exception of divalent iron, divalent tin, and trivalent antimony.

(5) The reaction is capable of detecting as little as 1 part of selenium in 38,000,000 parts of solution.

Having learned these properties of the reaction, attempts are now being made to discover an equation which represents the reaction between the ions. Attempts are also being made to apply this reaction to the quantitative determination of selenium in soils, plants, and steels.

Lactogenic Substance in Reptile Pituitaries. BERT CUNNINGHAM.

As is well known, the pituitary is the seat of secretion of a variety of endocrinal substances; the number accepted depending upon the school to which one belongs. There seems to be little doubt however as to the validity of prolactin.

Since the chemical formula of this substance has not been determined, and since the identity of the material in the turtle pituitary has not been established, it seems preferable to use the term lactogenic substance in this paper.

Prolactin is rather unique in that it produces responses of considerably different natures and upon different organs in birds and mammals. In pigeons and doves it affects the crop glands and in the latter the mammary gland, two structures which have nothing in common in origin. In the hen, which has neither the crop gland nor mammary glands, injection of prolactin produces, according to Riddle, a broodiness, which has led to the idea that it is a sort of maternal-instinct gland.

Since reptiles for the most part have no interest in their young, and since mating procedures are dependent upon secretions from gonad and pituitary (G. S. H.) it was thought worth while to look for prolactin in reptile pituitaries, and if found, to make more or less crude assays.

This has been done in five species of turtles, namely *Terrapene carolina*, *Chrysemys picta*, *Pseudemys troosti*, *Pseudemys hieroglyphica*, and *Pseudemys elegans*. The animals of the first two species were taken on

Long Island while the others were secured from the Carolina Biological Supply House. The pituitaries of the *T. carolina* were preserved in acetone for several weeks; those of the latter were either implanted immediately on removal or kept frozen until used.

Although the local stimulation technique may demonstrate smaller quantities of the factor, it seemed better to follow, in general, the crop-gland weight technique of Riddle in these experiments. Subcutaneous implants were made over the breast muscles. The results are shown in the accompanying table.

| SPECIES | PROLACTIN IN TURTLE PITUITARIES | | | | |
|-------------------------------|---------------------------------|----------------------------|--------------------|-------------|---------------------------|
| | Date taken | Average weight of implants | Number of implants | Test animal | Mg. of pituitary per unit |
| | | mg. | | | |
| <i>T. carolina</i> *..... | June | 3 | 5 | Pigeon | 1.0 |
| <i>C. picta</i> | Aug. | 5 | 4 | Dove | 5.0 |
| <i>P. hieroglyphica</i> | Sept. | 12 | 5 | Pigeon | 1.0 |
| <i>P. troosti</i> | March | 10 | 5 | Dove | 10.0 |
| <i>P. elegans</i> | March | 5 | 5 | Dove | 5.0 |

* Preserved in acetone.

While these assays are not as accurate as they might be if made on an "endocrine stock" such as used by Riddle they show quite clearly that prolactin does occur in assayable quantities in the pituitaries of chelonians.

At a later time E. M. Nixon and Martha Tanner, working in my laboratory, demonstrated the presence of a lactogenic substance in the pituitaries of the pine lizard, *Sceloporus undulatus*, and in the turtle *Emys blandingi*. They also repeated the assays of the box turtle and painted turtle pituitaries and secured results comparable to my previous assay.

The presence of a lactogenic substance in the pituitaries, therefore, has been demonstrated for six species of turtles and one lizard. The assays while crude show a considerable quantity of the factor in the glands of these reptiles.

The Nomenclature of Fractures. GROVER MURRAY.

The lack of existing specific fault, joint, and fracture cleavage terminology has instigated the writer to attempt a revision of these terms

without introducing an entirely new system of nomenclature. Contemporary usage involves a combination of structural, genetical, dynamical, and geometrical terms. Obviously, a dynamical or genetical term or classification cannot precede a structural, descriptive, or geometrical one. The writer suggests, therefore, that use be made at all times of: (1) descriptive and suggestive terminology, (2) dynamical terms only after the causal stresses have been determined, and (3) terms related to local movement on faults as defined by the Geological Society of America's Committee on Fault Nomenclature.

Descriptive terms have been applied to joints and to fracture cleavage, and modifications have been made in many of the fault definitions. The writer urges that the terms be used as defined therein and that their descriptive, structural, and geometric bases be retained.

Since the report of the Geological Society of America's Committee on the Nomenclature of Faults, professional writers have largely standardized many fracture terms. Other terms, however, have failed to become standardized. These disputed terms have been re-defined as specifically as possible. Existing, undisputed terms have been assembled for completeness.

On the Classification of Collineations. J. W. LASLEY, JR.

This paper presents a classification of non-singular collineations in the plane by reducing all cases to a common canonical form in which the matrix is that of Jordan. Attention is directed to the collineation itself rather than to derived properties of it which may or may not be applicable to a particular case. Assumptions as to incidence relations among the double elements are avoided. Instead of proving, as is customary, a number of theorems relative to incidence, one such theorem is proved; namely, that all non-singular collineations in the plane have at least one double line and at least one double point on it. With this as a point of departure all collineations regardless of the nature of the roots of the characteristic equation and of the rank of the characteristic matrix are put into one common form, which exhibits incidence relations among the double elements common to all cases. The matricial setting of the problem is emphasized. Assumptions as to rank fall naturally into place. A fuller use of duality is made than ordinarily. The presence and the effect of the background of linear substitutions is pointed out. Incidence properties of the various cases follow naturally from the canonical expressions. Although avoiding the theory of elementary divisors, the treatment is that of algebra, utilizing geometry.

It makes available for the student a simplified first approach to the theory by developing the fundamental unity back of seemingly diverse conditions. It makes possible an easy remembrance of the canonical forms for the separate cases.

Sets of Conjugate Matrices. E. T. BROWNE.

If M is a given square matrix of order n , and if M_1, \dots, M_{t-1} are $t-1$ matrices of the same order which possess the following properties:

- (1) The M 's are commutative in pairs;
- (2) If λ is a scalar and $f(\lambda)$ a scalar polynomial in λ such that the following identity holds:

$$(I) \quad (\lambda - M)(\lambda - M_1) \dots (\lambda - M_{t-1}) = f(\lambda),$$

then M_1, \dots, M_{t-1} are said to constitute a *set of conjugates* to M . If t is the smallest integer for which an identity of the type I holds, the set of conjugates is called a *reduced set*. Sets of conjugates have been studied by Taber, P. Franklin, E. Sokolnikoff, Pierce, Hermann and others. In this paper a thorough study is made of sets of conjugates and more general sets are obtained than those given hitherto. In particular, a special study is made of *cyclic* sets.

Chemical Analysis by Means of Infrared Spectra. E. K. PLYLER.

The line spectra of elements have been used to supplement chemical methods for the detection of the presence of elements. In the same manner infrared measurements can be used to detect the presence of molecules. Each molecule has a characteristic band spectrum. It has been found that an amount of 1/100 of one per cent water present in many organic liquids, such as the alcohols and acetone can be detected. By having a series of absorption curves for standard mixtures, the amount of water present in a liquid may be obtained. The work is being extended to include many different materials.

Cosmic Ray Showers Produced in Large Thicknesses of Various Materials.

KARL Z. MORGAN and W. M. NIELSEN.

The apparatus used in these experiments consists of several Geiger-Muller counter outfits; each adjusted to register only coincident discharges of three or more counters. The data were recorded by electric relays on a moving tape.

The first series of investigations was made on Beech Mountain (elev. 4540 ft.). The variation of doubles with grams/cm.² of lead above the

counters was found. An arrangement was used which gave an extremely low zero count of 6% of the maximum count. The experiments were repeated for iron absorbers. The maximums occurred at about 20 gm./cm.² for lead and 50 gm./cm.² for iron. The iron coincident counts at large thicknesses of scattering material were about 25% greater than for the lead.

A transition curve from iron to lead was obtained by placing various thicknesses of lead under 274.16 gm./cm.² of iron. This curve indicated an initial rise to 13.12 gm./cm.² of lead under the iron. Then there was a rapid decline in coincidences until about 100 gm./cm.² of lead was placed below the iron. Beyond this point the transition absorption curve followed the lead curve characteristic.

The 13.12 gm./cm.² of lead was placed above 274.16 gm./cm.² of lead and there was no increase in counting rate above the normal iron count. This seems to indicate a secondary multiplication process by which the heavier elements produce more cosmic radiation for softer producing radiation;—the mean free path is shorter in lead for soft producing radiation.

The 13.12 gm./cm.² of lead was placed below various thicknesses of iron. These points were plotted and connected and the general slope of this curve was the same as that of the iron curve.

Each material indicated a hard and a soft coefficient of absorption. The absorption coefficients of air radiation in lead were .03 cm²./gm. and .0006 cm²./gm.; of air radiation in iron they were .007 cm²./gm. and .00044 cm²./gm.; and of iron radiation in lead they were .009 cm²./gm. and .0006 cm²./gm. This seems to indicate a marked dependence of the absorption of this hard component of radiation upon mass.

The semi-log plot of the weak component of radiation was extrapolated and subtracted from the total coincident counts. This gave curves indicating second maxima of radiation. The second maxima were at about 200 gm./cm.² and 550 gm./cm.² for lead and iron respectively. The ratio of these two values was equal to the ratio of the gm./cm.² of the first maxima for these materials. This might indicate that the same mechanism could account for the production of showers by this extremely penetrating producing radiation as by the softer producing radiation.

Next a series of experiments, using two counter outfits running simultaneously, was made at Hickory, N. C. (elev. 1100 ft.). This gave a set of data showing the double, triple, and quadruple coincidences at various thicknesses of producing material. The quadruple maximum was

at a greater thickness than the triple or double maxima and there was a general symmetry between the three sets of curves obtained. This seems to be in further agreement with the multiplication theory of showers.

With a set of counters arranged to count doubles and 274.17 gm./cm². of lead above various thicknesses of iron a set of data was obtained. This gave an initial drop and then a rise to saturation with the iron curve as run at Hickory. This behavior may be explained by assuming a longer mean free path for radiation in iron. The rise to the iron curve is due possibly to the production of new shower producing radiation in the iron. All this evidence seems to be in agreement with the recently developed multiplication theory of showers.

Plastic Give and Recovery in Stretched Neoprene. MILTON L. BRAUN.

A band in the shape of a closed hairpin 7 cm. long, cut from a 2 mm. slab of vulcanized neoprene, was subjected to a constant tension of slightly more than a kilogram, or of 1.20×10^7 dynes per square centimeter of original cross section, in a chamber having controlled temperature in the range between 16 and 50 degrees Centigrade. The rate of change in length with change in temperature varied between -0.010 to -0.028 cm. per degree for nearly a year. A steady rate was found to give way to a very sudden increase in length when a certain temperature was attained. This was interpreted as a plastic give. Recovery from the plastic give was established on the new plane of length upon reduction of temperature, but the recovered temperature rate of change in length was always numerically greater than that before the give. These plastic excursions occurred at increasingly elevated temperatures. The duration of the stretch, the time of maintenance of a selected temperature, and the rate of change of temperature are factors which have not as yet been correlated with the plastic give or recovery. The specimen of neoprene was supplied through courtesy of the Rubber Laboratory of the E. I. DuPont de Nemours and Company, Incorporated.

H. L. BLUMQUIST, *Secretary.*

PROCEEDINGS OF THE ELISHA MITCHELL SCIENTIFIC SOCIETY

OCTOBER 13, 1936, TO MAY 11, 1937

370TH MEETING, OCTOBER 13, 1936

M. J. ROSENAU: *Epidemics of Influenza.*

A brief account of the history of influenza and its devastating pandemics which sweep the world at irregular intervals, about a generation apart. The mystery of the disease and recent advances concerning the discovery of its cause in the shape of a filterable virus. The modes of transmission, period of incubation, and other salient factors concerning the disease were given. Experiments with the disease on Cape Cod in the two towns of Brewster and Chatham were related and experiments during the great pandemics of 1917 and 1918 on the cause and mode of transmission of the disease were described.

371ST MEETING, NOVEMBER 10, 1936

S. E. SMITH: *Permeabilities of Cellulosic Films.*

The term "cellulosic films" includes films of regenerated cellulose and films of cellulose derivatives. The common property of the cellulose type "molecules" is vast length, relative to the other dimensions. Lateral attraction between these long chains results in the coherence of cellulose structures.

Cellulose films are hygroscopic. The moisture content is of prime importance to the condition of the film. The phenomena of swelling are well known among cellulose structures. The moisture content is also the determining factor in the permeability to gases. Dry cellulose films are completely impermeable to the permanent gases, while the permeabilities of moist films are in the ratio of the water solubilities of the diffusing gases.

T. F. HICKERSON: *Secondary Stresses in Bridge Trusses.*

When a bridge truss is subjected to primary axial stresses due to loads applied at the joints, the tension members become slightly longer, and the compression members correspondingly shorter. The

angles formed by the members meeting at a joint therefore tend to increase or decrease. This angular change may take place freely in the pin-connected truss without restraint, but if the truss members are rigidly connected at the joints due to riveting or welding, bending will be induced. The stresses resulting from this bending are called Secondary Stresses. They are greatest near the ends of the members.

The following steps indicate the general procedure in the proposed method for computing Secondary Stresses:

(1) Knowing the deformations in each member of the truss due to the primary axial stresses (obtained by dividing the products of the unit stresses times the length by the modulus of elasticity (E)), determine the displacements of the joints from a Williot diagram; or preferably, determine the rotations of the members from the computed angle changes of the truss triangles as though they were pin-connected.

(2) Determine the relative displacements (d/L -values) of the ends of each member from Step 1, with plus sign for clockwise rotation and minus sign for counter-clockwise rotation.

(3) Calculate each joint rotation (Θ) from the formula:

$$\Theta = \frac{3\Sigma K \frac{d}{L}}{2\Sigma K} \quad (1)$$

in which

$$K = \frac{I}{L} = \frac{\text{Moment of inertia}}{\text{Length of member}}$$

(4) Letting the letters N and F refer to the "near" and "far" ends of the members, calculate each joint rotation (Θ_N) by successive approximations; the first value is given by Eq. (1), and the other values are those brought over from the adjacent joints, the carry-over factor along any member being $-\frac{K}{2\Sigma K}$. Thus:

$$\Theta_N = \frac{3\Sigma K \frac{d}{L}}{2\Sigma K} - \frac{\Sigma K \Theta_F}{2\Sigma K} \quad (2)$$

(5) Compute the desired bending moments by means of the Slope-Deflection formula,

$$M_{NF} = 2EK \left(2\Theta_N + \Theta_F - 3E \frac{d}{L} \right) \quad (3)$$

from which the stresses follow directly.

372ND MEETING, DECEMBER 8, 1936

E. A. CAMERON: *On Loci Associated with Certain Osculants of a Plane Curve.*

Some geometric relations existing among certain osculants to a plane curve at a point were discussed. A study was made of the loci determined by these osculants as the point of contact is allowed to move along the base curve. Specialization of the base curve led to some interesting results.

J. F. DASHIELL: *The Experimental Background of the Gestalt Movement in Psychology.*

A few of the pioneer experiments leading to the development of the Gestalt school of psychological theory are described in simplified form. They are the phenomenon of apparent-movement (Wertheimer), the choosing of grays by hens, apes, and children (Köhler), the insightful learning of apes (Köhler). Other illustrations are incidentally introduced to clarify the Gestalt emphasis throughout upon wholeness as opposed to atomism in any field of psychology.

373RD MEETING, JANUARY 12, 1937

D. P. COSTELLO: *Notes on the Breeding Habits of Some Pacific Coast Nudibranchs.*

The nudibranchs are a group of marine gastropod molluscs characterized chiefly by the absence of a shell. In common with most gastropods, nudibranchs are hermaphroditic, and the animals, by pairs, practise reciprocal fertilization. Two to twenty-one days after mating, each member of the pair deposits a ribbon-like egg mass of fertilized eggs. The eggs are enclosed in capsules arranged in transverse rows across the ribbon, cemented together by a gel, and the ribbon is attached by one edge to the substratum in the form of a spiral of Archimedes. The egg ribbons of twenty species were studied and in all cases it was found that the spirals of Archimedes had been deposited in a counterclockwise direction (viewed dorsally).

OTTO STUHLMAN: *The Bio-Physics of the Human Ear.*

Published in the Jour. Amer. Acous. Soc. 9: 119. 1937.

374TH MEETING, FEBRUARY 2, 1937

EDWARD MACK, JR.: *Why Rubber Stretches.*

An attempt at a theory of the mechanism of elastic stretching in rubber was made. The explanation was given in terms (1) of the free

rotation of carbon atoms about single bonds, and (2) of the attraction between hydrogen atoms of the hydrocarbon molecules. Some of the more important properties of the various forms of rubber were discussed from the point of view of strict adherence to the principles of geometry and of structural organic chemistry. Among the topics discussed were: shape of the rubber molecule, the work of stretching rubber, the shape of the stress-strain curve, heat effects in rubber, and properties of synthetic rubbers.

375TH MEETING, MARCH 2, 1937

H. WARD FERRILL: *The Adrenals and Experimental Diabetes.*

To be published in full in *Archiv. of Int. Med.*

The etiology and course of diabetes mellitus has been associated with adrenal function by various authors, since Zuelzer (1907) proposed that a mutual antagonism exists between the adrenals and pancreas. Due to the disagreement among workers and due to the fact that the supposed relationship of the two glands has in recent times led to surgical procedures, this problem was undertaken and carried out on a suitably large number of animals to be of significance.

The report is concerned chiefly with experiments to determine whether reduction or suppression of epinephrine secretion from the adrenal glands exerts a significant influence on experimental diabetes. In these experiments the criterion was the amount of insulin required to maintain the level of sugar excretion in the urine below about five grams daily and the concentration of sugar in the urine below about one per cent. The experiments were performed on dogs.

All the experimental and control animals were kept under identical or comparable conditions, on a constant diet consisting of 500 grams of boiled beef lung and 100 grams of cane sugar daily, divided into two meals. In the morning meal 50 grams of fresh raw beef pancreas was added. Drinking water was available at all times. Daily administration of insulin was divided into two doses and given at the time of feeding. Urine was collected for 24 hours, the quantity excreted and the sugar content determined at the same hour each morning.

Interference with epinephrine secretion was accomplished by excision of one adrenal and denervation of the other gland. In addition to denervation, the medulla of the remaining gland was curetted by drilling with a dental burr, so that most of the medulla was destroyed or damaged. The adrenal operations in these animals were performed in two stages and pancreatectomy was performed during the interval between

the adrenal operations. Other dogs were depancreatized without subjecting the adrenals to operations for reducing the epinephrine secretion. These served as controls. At the end of different periods of observation the experiment was terminated and the animals sacrificed for quantitative determination of the degree of reduction or suppression of epinephrine secretion by the method employed by Stewart and Roff.

From this series of experiments it has been concluded that:

1. The supposed dependence of experimental pancreatic diabetes on epinephrine secretion from the adrenals is not supported by substantial experimental evidence.
2. Severity of diabetes in depancreatized dogs is not modified by reduction or suppression of epinephrine secretion from the adrenals.
3. Insulin requirement on a constant diet is within the same range in depancreatized dogs and in dogs with reduced or suppressed epinephrine secretion in addition to pancreatectomy.
4. Depancreatized dogs with reduced or suppressed epinephrine secretion are not more sensitive to insulin than ordinary depancreatized dogs.
5. Depancreatized dogs kept for some weeks on a constant diet with adequate amounts of insulin to control glycosuria sooner or later show a reduction or suppression of epinephrine output from the adrenals.
6. Results obtained on depancreatized animals that have been subjected to operations on the adrenals should be interpreted with extreme caution unless the animals are obviously in excellent physical condition.

W. C. GEORGE: *The Significance of Blood in the Classification of Animals.*

The cytological details of blood cells sometimes afford taxonomic details that may be of significant help in cases difficult of classification. In the ascidians there are certain features of blood histology that appear to be familial in distribution, others generic, and still others specific. For example, certain peculiar cells, called colorless morula cells, are present in all species of the family Ascidiidae studied; they are not known to occur in representatives of any other family. They have distinct features for the different species and subspecies known to the author. The genus *Ecteinascidia* appears to be distinguished, furthermore, from all other ascidians by the presence of fusiform granules in

the orange cells. These granules of orange pigment have characteristic size variations in the different species. The two genera of the family Botryllidae are distinguished from one another by the presence of pigmented cells characteristic of each. In addition to the colored cells common to both genera, the genus *Botryllus* has cells with dark blue pigment and the genus *Botrylloides* has cells with brownish red pigment.

376TH MEETING, APRIL 6, 1937

E. W. MCCHESENEY: *The Identification of the Amino Acids.*

Published in full in Jour. Amer. Chem. Soc. **59**: 1116, 1937.

T. F. HICKERSON: *Multi-Storied Building Frames.*

Using the Slope-Deflection equations for an exact solution of multi-storied frames involves the determination of the rotation of every joint and the sidesway of every story. Accordingly, one is confronted with as many equations, to be solved simultaneously, as there are joints and stories in the framework, unless the bent is symmetrical about a vertical center line, in which case the number of unknown rotations are reduced fifty per cent.

The formal algebraic solution becomes exceedingly laborious and time consuming when the number of equations exceeds three or four and therefore the so-called exact method is not accepted as a satisfactory office method.

Whatever procedure is adopted, an important equation of Statics is the following: "Total Shear in any Story, times Story Height, equals the sum of the Moments at the top and bottom of all the Columns of that Story."

Useful formulas will now be derived on the basis of (1) $\Sigma M = 0$ at a joint and (2) $\Sigma M = 0$ in a story. Let joint N of a building frame subjected to a horizontal sidesway (d) represent the "near" end of any four members having I/L - values of K, K₁, K₂, and K₃; and with corresponding "far" ends at joints F, F₁, F₂, and F₃.

Since the members at joint N are in equilibrium under sidesway, we may apply $\Sigma M = 0$ at the joint. Then substituting in the Slope-Deflection equations, noting that each member rotates through the same angle (Θ_N) at N, grouping terms, and solving, we have

$$\Theta_N = \frac{3\Sigma K \frac{d}{h} - \Sigma K \Theta_F}{2\Sigma K} \quad (1)$$

Dividing Eq. (1) in two parts, multiplying by E , and letting

$$x = E \frac{d}{h}$$

we have:

$$E\theta_N = \frac{3\Sigma Kx}{2\Sigma K} - \frac{\Sigma KE\theta_F}{2\Sigma K} \quad (2)$$

The first part of Eq. (2) is the total rotation at any joint (N) caused by the sidesway of the adjacent upper and lower stories as if the other ends were fixed; while the second part is the sum of the rotations brought over from the far ends of all the members meeting at joint N. It should be noted that the carry-over factor along any member is $-\frac{K}{2\Sigma K}$.

Assuming the rotation at the far ends equal, and the *same* as that at the near end (that is $\theta_F = \theta_N$); then Eq. (2) takes the form:

$$E\theta_N = \frac{\Sigma Kx}{\Sigma K} \quad (3)$$

If one member meeting at a joint is known to have its far end fixed, as at each of the upper joints of the first story of a frame fixed at the bottom, then Eq. (3) would be modified slightly, for these special cases.

(2) Let K, K' , etc. = $\frac{L}{L}$ — values of the columns in any story;

θ_U, θ_L , etc. = corresponding rotations at the upper and lower joints respectively;

d = sidesway of any story; that is, the sideswise deflection of upper floor relative to the lower;

$x = E \frac{d}{h}$ = sidesway quantity for that story;

M = story moment (but if frame is symmetrical about a vertical center line, M = one-half "total shear in story" times "story height" and only one-half of the columns are considered); and $m = \frac{1}{2}M$.

If A, B , etc. are the columns in the story, we have:
for Column A,

$$M_U = 2 EK \left(2\theta_U + \theta_L - 3 \frac{d}{h} \right).$$

and

$$M_L = 2 EK \left(2\theta_L + \theta_U - 3 \frac{d}{h} \right)$$

for Column B,

$$M'_U = 2 EK' \left(2\theta'_U + \theta'_L - 3 \frac{d}{h} \right)$$

and

$$M'_L = 2 EK' \left(2\theta'_L + \theta'_U - 3 \frac{d}{h} \right); \text{ etc.}$$

Adding the foregoing expressions, and applying $\Sigma M = 0$ to the columns in the story, we have

$$6K(E\theta_U + E\theta_L) + 6K'(E\theta'_U + E\theta'_L) - \left(6E \frac{d}{h} \right) [2(K + K' + \dots)] + M = 0$$

from which

$$x = E \frac{d}{h} = \frac{m + \Sigma K(E\theta_U + E\theta_L)}{2 \Sigma K} \quad (4)$$

In order to avoid possible confusion, it should be seen that Eqs. (1)-(3) include the K 's of the members meeting at a joint; while Eq. (4) refers only to the K 's of the columns in any story.

Label the joints and stories in consecutive order from the bottom upwards. Thus, let x_1 equal the sideways quantity in the first story, x_2 equal that in the second story, etc.

The following steps are to be taken in carrying out the proposed method of analysis.

(1) Use Eq. (3) for finding the joint rotations in terms of the x 's of the adjacent stories; set down the results in tabular form.

(2) Carry over the rotations from the far ends to the near end, using the factor $-\frac{K}{2\Sigma K}$.

(3) Apply Eq. (4) to each story and solve for x .

Note: Assuming as a first approximation, all the x 's above any given story to be equal, not more than two equations need be solved simultaneously at a time even for multi-storied frames.

(4) Knowing x for each story the joint rotations can be found by successive approximations and finally the desired bending moments can be calculated from the formula,

$$M_{NF} = -2K(3x - 2\theta_N - \theta_F) \quad (5)$$

As a final check on the calculations, the girder moments at a joint should equal the sum of the column moments at that joint.

377TH MEETING, MAY 11, 1937

R. F. POOLE: *Progress in the Control of Plant Diseases.*

The cryptogamic botanist and mycologist first conceived the possibilities of controlling organisms that cause plant diseases. Since the beginning of the Land Grant Colleges the plant pathologist has contributed much to the practical development of quality products and an economic agriculture. Plant pathology aims specifically toward economic and quality production rather than toward quantity production. The measures used for the control of plant diseases were discussed under six headings; namely, (1) Resistant varieties, (2) Rotation of crops, (3) Soil treatment, (4) Adaptability, (5) Sanitary practices, and (6) Chemical treatments. Important advancements in technique, in the perfection of chemical formula and application, the behavior of organisms in changed soil media and the destruction of inoculum were discussed. The lecture was illustrated, demonstrating the serious losses caused by plant diseases and the effects of practical control measures.

The following officers were elected for the year 1937-1938:

President—John N. Couch.

Vice-President—H. D. Crockford.

Secretary-Treasurer—E. L. Mackie.

The permanent secretary, E. T. Browne, and the editors of the Journal, W. C. Coker, Otto Stuhlman, and C. D. Beers, the last-named having been elected at the December meeting to fill the vacancy caused by the resignation of Dr. H. V. Wilson, were continued in office.

A CLASSIC PROBLEM IN EUCLIDEAN GEOMETRY

A BASIC STUDY*

By ARCHIBALD HENDERSON

I

For approximately a century the harmless-looking exercise in Euclidean geometry, studied in the present monograph, has attracted from mathematicians a measure of attention apparently disproportionate to its importance. Among the many who have attacked it may be mentioned Todhunter, Sylvester, Greenstreet, Lemoine, Meurice, Tarry, Lanvernay, Halsted, Birkhoff, and Hopkins. Numerous solutions, some authentic but many the reverse, have been published in England and the United States and on the continent of Europe. It is not a little singular that the published "solutions" are, in a surprising number of cases, incomplete, inadequate, incorrect, and even grossly and ludicrously erroneous.¹

Despite the appreciable number of isolated "solutions" which have, often incautiously, been submitted to public scrutiny, no thoroughgoing analysis of the problem has ever been made. It is primarily on this account that I have ventured upon this diverting excursion and pleasure trip. Indeed I may wellnigh venture to describe this paper as an essay on the internal bisector problem to end all essays on the internal bisector problem. In 1900 one G. D. Birkhoff, then unknown to

* This study is dedicated to George D. Birkhoff, President of the American Association for the Advancement of Science. Cf. *infra*.

¹ For example, the solution proposed by the Rev. William Mason and endorsed by T. T. Wilkinson is unsound, as is also the discussion by the latter. See *The Lady's and Gentleman's Diary*, No. 156, pp. 87-8 (1859), and No. 157, pp. 84-86 (1860). It seems incredible that the erroneous "solution," submitted by F. G. Hesse, should ever have been recommended by J. J. Sylvester and N. M. Ferrers as "thoroughly sound" and actually published in the *Philosophical Magazine*, 4th ser. 47: 354-6 (1874). Nine different purported solutions appeared in the *American Mathematical Monthly* 2: 157-8, 189-92; 5: 108-9; 7: 226-8; but the mortality was devastating. Only two of the solutions were free from flaws, one of those being indirect, the other by trigonometry. Even the "solution" by the editor, admittedly "adapted" from a solution by the editor of the mathematical department of *The Educational Times*, is unsound.

fame, submitted to the editor of the *American Mathematical Monthly* a solution of the proposed "teaser," a solution which was not published, Birkhoff's name being included in the list of "others who submitted solutions." As almost all the published "solutions" were absurdly erroneous, the editor might perhaps better have taken a chance on publishing Birkhoff's solution, now irrevocably lost, than those he was so injudicious as to print, including his own erroneously "adapted" one. A certain piquancy attaches to the task of making an exhaustive study of this problem and its history. It is like breaking a butterfly on a wheel. And yet—the Biblical parable of the grain of mustard seed or the familiar rhyme about the little acorn finds convincing exemplification here. Felix Klein wrote delightfully and searchingly of three famous geometrical classics of antiquity: the squaring of the circle, the duplication of the cube, and the trisection of the angle. David Hilbert delivered some charming lectures on the foundations of geometry. Einstein and Birkhoff divided honors over the mathematical recreation of the impact of two billiard balls according to the mechanics of special relativity. With such stimulating examples of recreational relaxation before me, I see no bar to vying with them in the field of mathematical diversion, and by searching analysis magnifying an innocent-looking geometrical exercise into a classic of modernity.

II

In the course of these researches, which have led me to many new proofs, I have discovered that this little exercise, shy and shrinking as the modest violet, has wide ramifications of surprising interest and variety. Invoked for its proof are principles of continuity, symmetry, inversion, reciprocation, antiparallels, cross ratio, harmonic ranges and pencils, the complete quadrangle, the question of constructibility by means of ruler and compasses. The exercise seems to stand on the border-line between metrical and projective geometry, between Euclidean and non-Euclidean geometry, sharing alike the adventitious yet highly effective aid of analytic geometry and higher plane curves. We need not be surprised to discover that the exercise is a special case, one of an infinite number of similar special cases, of a theorem of a more general nature. Study of this more general theorem enables us to view the whole problem in a new perspective, giving rise as it does to provocative questions regarding other interesting features of, and configurations associated with, the triangle.

It is passing strange that this problem, so far as I have been able to

discover, received no consideration or even mention by geometers, ancient or modern, in either direct or converse form, until about a century ago. Four issues are involved: (a) to find new direct proofs by theorems in the books which succeed Book I of Euclidean geometry as we have it today; (b) to find new indirect proofs; (c) to find that which not a few mathematicians have failed to find, direct proofs by the use of theorems in Book I only; and (d) to clear up the obscurity and mystery concerning the problem's exaggerated yet unmistakably cryptic difficulties.

These difficulties are all the more surprising in view of the fact that the problem under consideration is the converse of an exercise in Book I, the proof of which offers no difficulties whatever.² The study of the converse of this almost trivial exercise has thrown into sharp relief several theorems, commonly slighted or wholly ignored in works on geometry ancient or modern, which are nevertheless of genuine interest and fundamental importance. The present treatment of the internal bisector problem, with its many implications and ramifications, may perhaps furnish fresh information and new points of view, even to the trained mathematician.

III

In a French journal,³ just ninety-five years ago, was proposed, for the first time so far as is at present known, the following problem under the caption "Théorèmes à Démonstrer.—Problèmes":

THEOREM 1. *To demonstrate that if in a rectilinear triangle two interior angular bisectors are of equal length, the triangle is isosceles.*

This is the converse of the very easy exercise: "The internal bisectors

² The writers of textbooks in geometry almost invariably omit the problem of this paper. Considerable space with adequate references, however, is accorded it in J. S. Mackay's *Euclid*; and a solution by Descube is given in Mackay's *A Key to Euclid*. The exercise is assigned in George C. Edwards's *Elements of Geometry* (1896), being included among the miscellaneous exercises on page 138. In G. A. Wentworth's *Plane Geometry* (revised edition, 1899), p. 72, the converse of our problem is assigned and its "opposite" is solved in the text; but the pupil is asked, not to solve, but merely to state, our problem and to tell whether it is true. In the same author's *Exercises in Wentworth's Geometry* (revised edition, 1880), p. 10, the converse of our problem is proved; but our problem and its opposite are merely stated without comment. No reference to this problem, rather surprisingly, is found in Julius Peterson's *Methods and Theories for the Solution of Problems of Geometrical Construction* (English translation, 1923), a work almost more difficult to read in the weird translation than in the original Danish (1866).

³ *Nouvelles Annales de Mathématiques* 2: 57 (1842).

of the base angles of an isosceles triangle, terminating in the sides including the third angle, are equal." Soon after the appearance of the problem to be solved, two solutions appeared in the same journal of the same year, one of them indirect and each using theorems of Books II and III, in particular the same somewhat unfamiliar theorem which is fundamental in the theory of inversion.⁴

It was not until almost ten years later that the problem appears to have piqued the interest of mathematicians of the University of Cambridge, where, as Sylvester observes, "it excited considerable attention among the mathematicians of the place." Certainly Isaac Todhunter did not shine in this affair, as he failed to obtain a direct solution; and Sylvester also betrayed himself as surprisingly inept in his approach to the entire problem. In the first paper which appeared in English on the subject, Sylvester, then of the Royal Military Academy, Woolwich, erroneously conceived the notion that the problem was incapable of direct demonstration. He ventured to indulge in the following bantering comment, presumably aimed at Dr. Isaac Todhunter of Cambridge:

If report may be believed, intellectuals capable of extending the bounds of the planetary system and lighting up new regions of the universe with the torch of analysis, have been baffled by the difficulties of the elementary problem stated at the outset of this paper, in consequence; it is to be presumed, of seeking a form of geometrical demonstration of which the question from its nature does not admit. If this be so, no better evidence could be desired to evince the importance of such a criterion as that suggested in the text.⁵

The criterion suggested is thus stated by Sylvester: "Whenever the truth of a geometrical theorem depends on the necessary non-existence of real roots (between prescribed limits) of the analytical equation ex-

⁴ Solution by M. Rougevin, "élève de la classe de Mathématiques élémentaires du Collège Louis-le-Grand [Institution Lorial]", *ibid.*, 1: 138-9; and solution by Mr. Grout de Saint-Paer, "élève du Collège de Versailles," i, 311. The solution by T. T. Wilkinson, F. R. A. S. in *The Lady's and Gentleman's Diary*, No. 154, pp. 58-9 (1856) is virtually identical with that of Rougevin and presumably based upon it.

⁵ On a Simple Geometrical Problem illustrating a Conjectured Principle in the Theory of Geometrical Method. *Philosophical Magazine* 4: 366-9 (1852). The same article is found in Sylvester's *Collected Works* 1: 392-5 (1837-1853). One may observe that no better evidence than this could be desired, that the difficulties which Todhunter, Sylvester, and other English mathematicians could not overcome, in the effort to find a direct demonstration of the problem, were very real difficulties indeed.

pressing the conditions of the question, no other form of proof than that of the *reductio ad absurdum* is possible in the nature of things." This criterion was advanced by Sylvester because he was unable to devise a direct proof of a theorem upon which the bisector problem may be shown immediately to depend, namely:

THEOREM 2. *If from the middle of a circular arc two chords of the circle be drawn, the remoter segments of the two chords cut off by the chord of the given arc being equal, then the nearer segments will also be equal.*

Referring to this theorem, Sylvester says: "As an example, I throw out (not a challenge, but) an invitation to discover a direct proof, if such exist, of the following geometrical theorem, as simple a one as it is perhaps possible to imagine. . . ."

IV

As Theorem 2 is of some intrinsic interest as well as the basis of several proofs of the bisector problem, I present below a simple proof unlike others I have seen. The theorem is one of a group of associated theorems, all capable of similar direct demonstrations. Taken together they constitute the fundamental principle of the theory of inversion.

Given N the mid-point of circular arc BNC ; and NK_1A_1 , NK_2A_2 two chords, with $K_1A_1 = K_2A_2$ (Fig. 1). From N drop a perpendicular NM on chord BC , meeting the circle again in L ; and let the chord LA_1 produced meet BC produced in D . Join N , A_1 , A_2 by chords to B and C . From the pairs of congruent triangles MNK_1 , A_1NL and MNK_2 , A_2NL , we have

$$NL \cdot NM = NK_1 \cdot NA_1 = NK_2 \cdot NA_2 \dots \dots \dots (1)$$

Hence

$$NK_1(NK_1 + K_1A_1) = NK_2(NK_2 + K_2A_2)$$

giving $NK_1 = NK_2$, since $K_1A_1 = K_2A_2$ by hypothesis. We may note, therefore, that $MK_1 = MK_2$ and $\angle A_1K_1C = \angle A_2K_2B$. Moreover,

* This invitation evoked a reply, in three parts, by James Adamson, D.D., which while very diffuse contains a satisfactory answer to Sylvester's query, together with a refutation of his unsound criterion: "On a proposed Test of the Necessity of Indirect Proof of Geometrical Demonstrations, with Remarks on Methods of Demonstration." *Philosophical Magazine* 1: 297-9, 332-8, 405-410 (1853). Other, chiefly brief, contributions to the subject appeared in *The Educational Times*, 1853 *et seq.*, *passim*. An extremely lengthy and complicated proof of the bisector problem, by means of reciprocation and the use of the external bisectors, is that of T. K. Abbott, *Philosophical Magazine* 5: 286-7 (1853).

from the pairs of similar triangles MNB, MBL and MNC, NCL, we have

$$\overline{NB}^2 = \overline{NC}^2 = NL \cdot NM \dots \dots \dots (2)$$

Hence from (1) and (2),

$$\overline{NB}^2 = \overline{NC}^2 = NK_1 \cdot NA_1 = NK_2 \cdot NA_2$$

which is the fundamental equation of inversion $\rho\rho' = a^2$. Furthermore, A_1N , A_2N are the internal bisectors of angles BA_1C , BA_2C , since

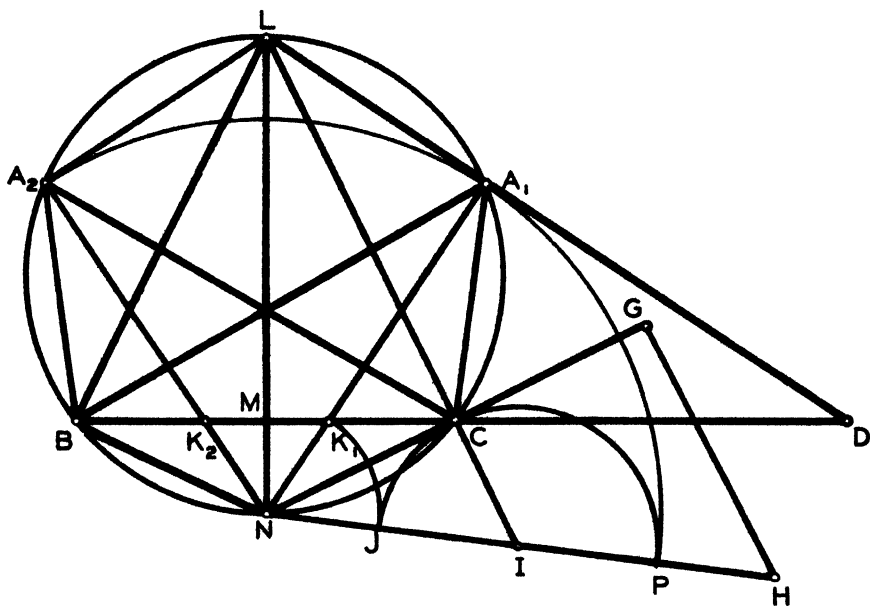


FIG. 1

each of the four angles BA_1N , CA_1N , BA_2N , CA_2N is measured by one-fourth the arc BNC. Since this is also the case with angles BCN, CBN, we may also derive equation (3) by comparison of the two pairs of similar triangles NK_1C , NCA_1 and NK_2B , NBA_2 . We should also note that LD is the external bisector of $\angle BA_1C$ since LA_1 is perpendicular to NA_1 , and that B, K_1 , C, D form a harmonic range, giving the proportions $BK_1 : K_1C :: K_2C : BK_2 :: A_1B : A_1C :: A_2C : A_2B :: BD : DC$.

Since from equation (1), if $NK_1 = NK_2$, we have $NA_1 = NA_2$ and

$K_1A_1 = K_2A_2$; and since if $NA_1 = NA_2$, we have $NK_1 = NK_2$ and $K_1A_1 = K_2A_2$, we may formulate two more theorems:

THEOREM 3. *If from the middle of a circular arc two chords of the circle be drawn, the nearer segments of the two chords cut off by the given arc being equal, the remoter segments will also be equal.*

THEOREM 4. *If from the middle of a circular arc two equal chords of the circle be drawn, the nearer and the remoter segments of the two chords cut off by the chord of the given arc will be equal, each to each.*

By a similar argument we may readily demonstrate the following theorem:

THEOREM 5. *If, from a fixed point (N) on the perpendicular bisector (NL) of a given segment (BC), a line be drawn within the angle CNL cutting BC at K_1 and determining A_1 from the relation $\overline{NC}^2 = \overline{NK_1} \cdot \overline{NA_1}$, then B, N, C, A_1 are concyclic.*

Thus

$$\overline{NK_1}(\overline{NK_1} + \overline{K_1A_1}) = \overline{NC}^2 = \overline{NM}^2 + \overline{MC}^2 = \overline{NK_1}^2 - \overline{MK_1}^2 + \overline{MC}^2$$

Hence

$$\overline{NK_1} \cdot \overline{K_1A_1} = \overline{MC}^2 - \overline{MK_1}^2 = (\overline{MC} + \overline{MK_1}) \cdot (\overline{MC} - \overline{MK_1}) = (\overline{BM} + \overline{MK_1})(\overline{MC} - \overline{MK_1}) = \overline{BK_1} \cdot \overline{K_1C}$$

Therefore B, N, C, A_1 are concyclic.

The results of Theorems 2, 3, 4 and 5 may be stated in the following form:

THEOREM 6. *Two triangles (A_1BC , A_2BC) having the base (BC), vertical angle (BA_1C , CA_2B) and bisector of the vertical angle (A_1K_1 , A_2K_2) equal, each to each, are congruent.*

This follows directly from symmetry about the line NML. It is extraordinary that this basic theorem regarding two triangles is conspicuous by its absence from virtually all text books and treatises on geometry in any language. If one triangle A_1BC is in position, the other may immediately be constructed by determining A_2 as the intersection of the circle through A_1 , C, N, B with the circle of center N and radius NA_1 . Theorem 6 may be stated as a problem in construction.

CONSTRUCTION 1. *To construct, on BC as a base and on the same side of BC, a triangle congruent to $\triangle A_1BC$: find the intersection of the circumscribing circle of the $\triangle A_1BC$ and the circle of center N and radius NA, where N is the meet of the perpendicular bisector of BC with the circum-circle BCA_1 . The point thus found (A_2) is the vertex of the required triangle A_2BC .*

We are now in a position to formulate two constructions for the following problem in construction: *Given the parts a , A , t_a of a triangle ABC ; to construct the $\triangle ABC$.*

CONSTRUCTION 2. At C lay off below BC ($= a$) an angle equal to $\frac{1}{2}A$, the other leg cutting the perpendicular bisector of BC at M , in N (Fig. 1). Let the circumcircle of $\triangle BNC$ cut NM in L . If we imagine the required triangle drawn in the position A_1BC , we have

$$NK_1 \cdot (NK_1 + t_a) = \overline{NC}^2$$

giving

$$NK_1 = \frac{1}{2}(-t_a + \sqrt{(t_a)^2 + 4\overline{NC}^2})$$

This can readily be constructed by extending NC to G , making $CG = NC$, and erecting a perpendicular to NG at G , laying off on it $GH = t_a$. Join H to N , the line HN cutting the perpendicular to NG at C in I . Then

$$NI = \frac{1}{2} \sqrt{(t_a)^2 + 4\overline{NC}^2}$$

Laying off IJ toward N equal to $\frac{1}{2}t_a$, we have

$$NJ = \frac{1}{2}(-t_a + \sqrt{(t_a)^2 + 4\overline{NC}^2}) = NK_1$$

Hence with N as center and NJ as radius describe arc cutting BC at K_1 . Then NK_1 extended cuts the circle BNC in the vertex (A_1) of the required triangle.

CONSTRUCTION 3. For NK_1 we may write $NA_1 - t_a$ and therefore $(NA_1 - t_a) \cdot NA_1 = NC$, giving

$$NA_1 = \frac{1}{2}(t_a + \sqrt{(t_a)^2 + 4\overline{NC}^2})$$

Lay off, on NI extended, $IP = IC$, giving

$$NP = \frac{1}{2}(t_a + \sqrt{(t_a)^2 + 4\overline{NC}^2}) = NA_1$$

The circle with N as center and NP as radius will cut the circle BNC in the vertex (A_1) of the required triangle.

PROOF I. The bisector problem is immediately solved by means of Theorem 6. Given BE , CF the equal bisectors of the base angles B , C of $\triangle ABC$ meeting in O (Fig. 2). Then AO is the bisector of $\angle A$, since

the internal bisectors of the three angles of a triangle are concurrent. Hence $\triangle ABE = \triangle ACF$, by Theorem 6, and therefore $AB = AC$.⁷

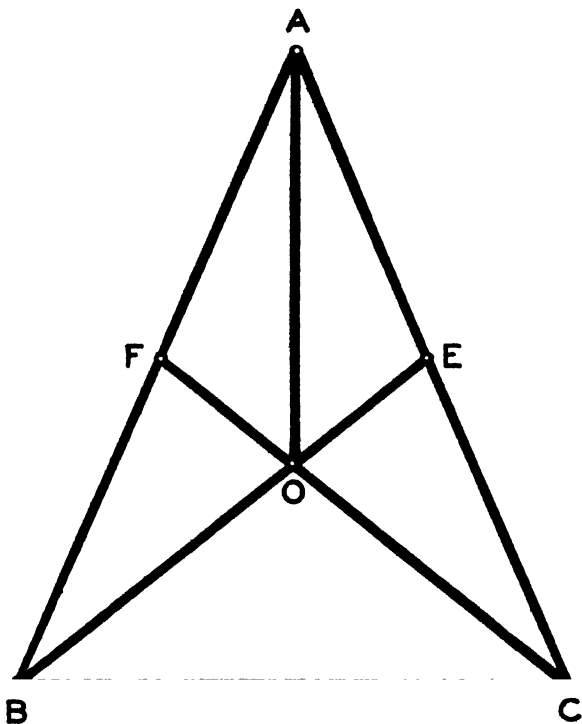


FIG. 2

V

In this section will be given seven new direct proofs of the bisector problem, based upon theorems in Books II and III. The first four of these proofs, succinct and elegant, are intimately connected.

PROOF II. Given $\triangle ABC$ with AD internal bisector of $\angle BAC$ (Fig. 3). At C lay off from AC and in the same sense an angle equal to $\angle ADE$, the other side meeting AD produced in L . Hence

$$AB \cdot AC = AD \cdot AL = AD(AD + DL)$$

giving

$$AD^2 = AB \cdot AC - AD \cdot DL \dots \dots \dots (1)$$

⁷ With a figure similar to Fig. 1, Rougevin, *l.c.*, gives an indirect proof, involving comparison of arcs, whereas the above proof is direct.

Since $\triangle CDL$ is similar to $\triangle ADB$,

$$AD \cdot DL = BD \cdot CD \dots \dots \dots (2)$$

From (1) and (2) we have

$$\overline{AD}^2 = AB \cdot AC - BD \cdot CD \dots \dots \dots (3)$$

Now from the bisector property, we have

$$AB \cdot CD - AC \cdot BD = 0 \dots \dots \dots (4)$$

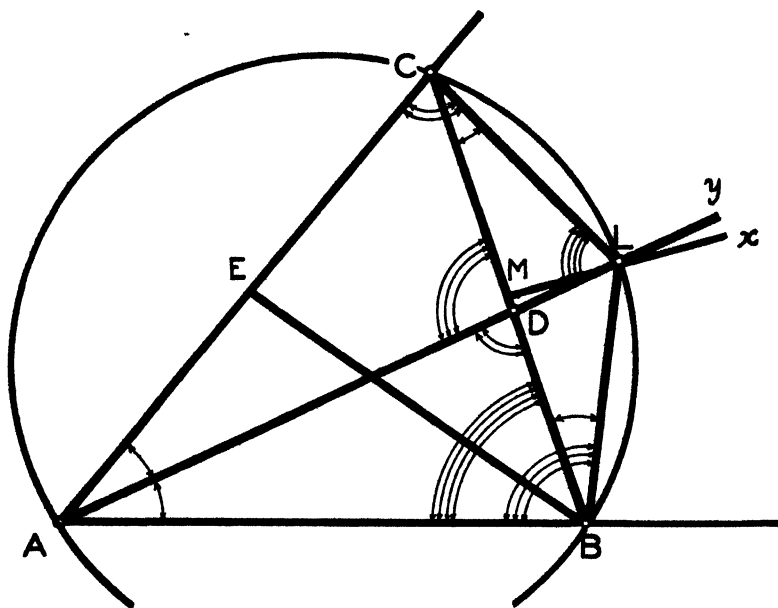


FIG. 3

Adding (4) to, and subtracting (4) from, (3), we have the familiar forms:

$$\overline{AD}^2 = (AC + CD)(AB - BD) \dots \dots \dots (5)$$

and

$$\overline{AD}^2 = (AB + BD)(AC - CD) \dots \dots \dots (6)$$

From (4) the right hand sides of equations (5) and (6), it may be noted, are equal, by composition and division:

$$AB + BD : AB - AD :: AC + CD : AC - CD \dots \dots \dots (7)$$

The proof now follows at once. If BE is the internal bisector of $\angle ABC$, we have, after the analogy of (5) or (6),

$$\overline{BE}^2 = (BC + CE)(AB - AE) \dots \dots \dots (8)$$

Hence if $AD = BE$, we have from (5) and (8),

$$\frac{BC + CE}{AC + CD} = \frac{AB - BD}{AB - AE} = \frac{AB - (BC - CD)}{AB - (AC - CE)} =$$

$$\frac{AB + CE + CD}{AB + CE + CD} = 1 \dots \dots (9)$$

by combining first and third fractions. Hence

$$BC - CD = AC - CE \text{ or } BD = AE$$

Since $\triangle ABE = \triangle ABD$, $\angle BAC = \angle ABC$, giving $BC = AC$.⁸

PROOF III. We may also proceed as follows: From AB and in the same sense lay off an angle equal to $\angle ADC$, the other side meeting AD produced in K say (not shown in Fig. 3, but identical with L). The remainder of the proof is precisely analogous to Proof II.

PROOF IV. Draw the circumcircle of $\triangle ABC$ cutting AD produced at L. The proof follows as above.

PROOF V. Erect perpendicular bisector of BC, at its mid-point M, meeting AD produced at L. If circumcircle of $\triangle ABC$ meets AD produced at K, then K coincides with L, since $LB = LC$ and chord KB = chord KC. Then since $\triangle ACL$ is similar to $\triangle ADB$, $AD + DL : AC = AB : AD$. The remainder of the proof follows as in Proof II.

PROOF VI. The next proof utilizes the principle of inversion and brings out a property discovered by Lanvernay and later arrived at independently by Meurice.⁹ The construction of Fig. 4 was suggested by equation (6) in Proof II above:

$$\overline{AD}^2 = (AB + BD)(AC - CD)$$

A similar proof, with figure to correspond, may also be derived from equation (5), Proof II.

Given $\triangle ABC$ and AD bisector of $\angle BAC$ (Fig. 4). Lay off from C

⁸ A more lengthy and cumbersome treatment is that of W. E. Heal, *American Mathematical Monthly* 24: 344-5.

⁹ E. Lanvernay, *Mathesis* 14: 40 (1894). In the same volume of the same journal, page 92, M. L. Meurice gives a proof of Lanvernay's Theorem by the use of an escribed circle.

toward A, $CH = CD$, and join H to D. Lay off on AB toward B, $AG = AH$; and from A drop a perpendicular AI upon DG extended to J, laying off $IJ = DI$. Let circle through D, A, J meet AB produced at K, and join K to D. Then

$$\overline{AD}^2 = AK \cdot AG \dots\dots\dots (1)$$

by the principle of inversion. Since also $AG = AH$ and $\angle HAD = \angle DAK$, $\triangle AHD$ is similar to $\triangle ADK$ and

$$\angle ADH = \angle AKD \dots\dots\dots (2)$$

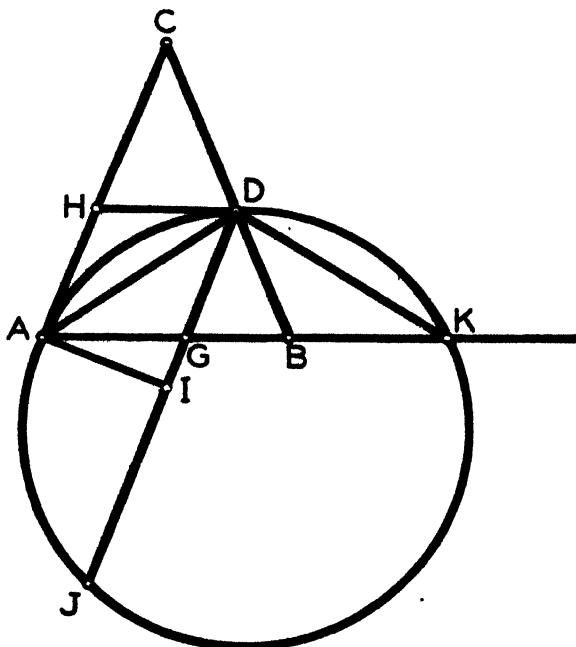


FIG. 4

Now $\angle DGK + \angle GDB + \angle BDK + \angle AKD = 180^\circ$ and $\angle CDH + \angle ADH + \angle ADG + \angle GDB = 180^\circ$. Since $\angle CDH = \angle CHD = \angle DGK$, and $\angle ADH = \angle ADG = \angle AKD$ by (2), we have $\angle BDK = \angle ADG = \angle ADH = \angle AKD$ by (2) and

$$BD = BK \dots\dots\dots (3)$$

Hence

$$\overline{AD}^2 = AK \cdot AH = (AB + BK)(AC - CH) = (AB + BD)(AC - CD)$$

The remainder of the proof follows as in Proof II.

PROOF VII. Given $\triangle ABC_1$, with $\angle AC_1B = \angle C$ (Fig. 5). The bisectors AD_1 , BE_1 of the angles BAC_1 , ABC_1 meet in the point P_1 . Prolong $C_1P_1F_1$, to meet the perpendicular bisector of AB at M , in the point N ; and draw the straight line BN . Now the circumcircle of the $\triangle ABC_1$

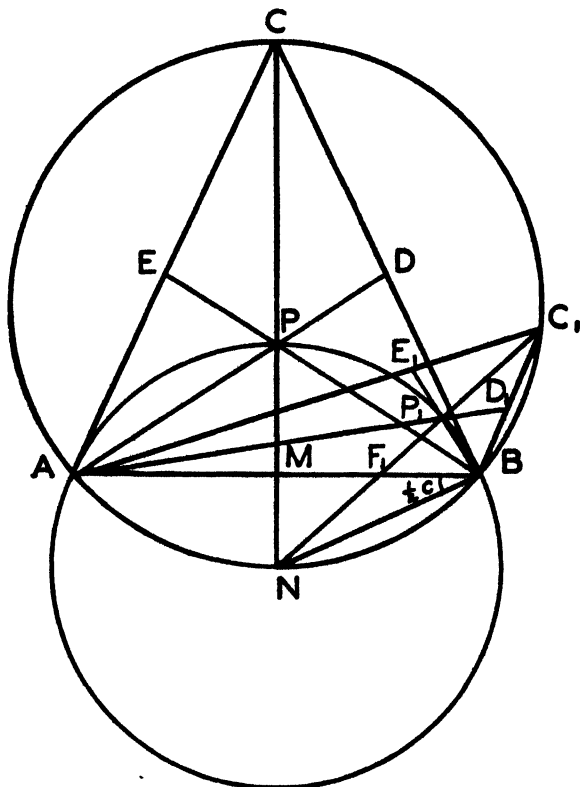


FIG. 5

passes through N , since CP_1 meets it in the mid-point of arc AB , which lies on the line MN . Hence

$$\angle ABN = \angle AC_1N = \frac{1}{2} \angle AC_1B = \frac{1}{2} \angle C$$

Now

$$\angle AP_1F_1 = \frac{1}{2} \angle BAC + \frac{1}{2} \angle AC_1B$$

and

$$\angle BP_1F_1 = \frac{1}{2} \angle ABC_1 + \frac{1}{2} \angle AC_1B$$

Therefore

$$\begin{aligned} \angle AP_1B &= \frac{1}{2} \angle BAC_1 + \frac{1}{2} \angle ABC_1 + \angle AC_1B = 90^\circ - \\ &\quad \frac{1}{2} \angle AC_1B + \angle AC_1B = 90^\circ + \frac{1}{2} \angle C \end{aligned}$$

which is a constant angle. Now describe a circle with center N and radius $NA = NB$, cutting NM produced in P. Then

$$\angle NPB = \angle NBP = 45^\circ + \frac{1}{2}C$$

since $\angle MNB = 90^\circ - \frac{1}{2}C$. Prolong NMP to meet the circle through A, N, B, and C_1 in C; draw straight lines AC and BC, and prolong AP, BP to meet BC, AC in D, E respectively. Since $\angle AP_1B = \angle APB = 90^\circ + \frac{1}{2}C$, P_1 describes the arc BP_1P as C_1 describes the arc BC_1C .

Let us now recall the theorem: "In every triangle ABC, to a greater side AC is drawn a smaller internal bisector BE" (For a proof of this theorem, cf. E. Catalan, *Théorèmes et Problèmes de Géométrie Élémentaire*, Theorem VIII (6th edition, revised and enlarged. Paris. Dunod. 1879).¹⁰ As C_1 traverses arc from B to C, AC_1 is always greater than BC_1 , since arc ACC_1 is greater than arc BC. Hence AD_1 for this interval is always greater than BE_1 . Similarly, for the symmetrical arc AC, BE_1 remains greater than AD_1 . When C_1 reaches C and P_1 reaches P, then D_1 reaches D and E_1 reaches E. Now AD, BE are the bisectors of the angles BAC, ABC respectively. But $\angle PAB = \angle PBA$, since $AP = BP$. Now $\triangle APE = \triangle BPD$, since $AP = BP$, $\angle APE = \angle BPE$, and $\angle PAE = \angle PBD$. Therefore $PD = PE$, and consequently $AD = BE$. Note that this is otherwise obvious, if we invoke the principle of continuity. Since C_1 is at C when P_1 is at P, $AC = BC$.

This is perhaps the most satisfying of all the proofs, since the internal bisector problem and its converse are both seen to be true, by the use of this figure.

PROOF VIII. Given $\triangle ABC$ with equal internal bisectors BE, CF of base angles ABC, ACB (Fig. 6). Lay off $\angle CFx = \angle EBA (= \frac{1}{2} \angle B)$, and $\angle FCy = \angle BEA (= \frac{1}{2} \angle C)$, Fx and Cy meeting at A_1 . Then $\triangle FCA_1 = \triangle BEA$. Hence the bisectors of $\angle FAC$ and $\angle FA_1C$

¹⁰ This theorem immediately furnishes a solution of the internal bisector problem, by the indirect method.

meet at N , mid-point of arc FNC of circle passing through F , N , C , A_1 , A . Let NA , NA_1 meet FC in K and K_1 ; then $AK = A_1K_1$, being corresponding parts of congruent triangles ABE and A_1FC . Now

$$\angle FCN = \angle CFN = \frac{1}{2} \angle FAC = \frac{1}{2} \angle FA_1C,$$

each being measured by one-half of arc FNC . Then NF , NC are tangents to circumcircles of $\triangle AKF$, $\triangle A_1K_1C$ respectively, giving $\overline{NF}^2 = NK \cdot NA$ and $\overline{NC}^2 = NK_1 \cdot NA_1$. Hence $NA = NA_1$ and $NK = NK_1$, since $NF = NC$ and $KA = K_1A_1$. Then $\angle NAA_1 = \angle NA_1A$; and since $\angle NAC = \angle NA_1F$, we have $\angle CAA_1 = \angle FA_1A$. Conse-

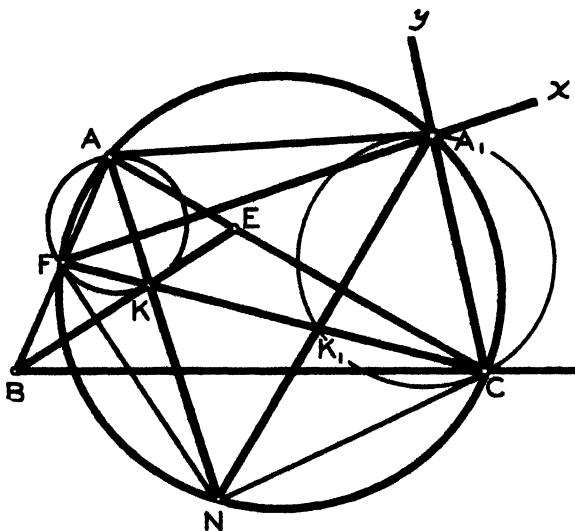


FIG. 6

quently arc $AF = \text{arc } A_1C$, and chord $AF = \text{chord } A_1C = AE$. Therefore $\triangle AFK = \triangle AEK$, and consequently $\triangle AEB = \triangle AFC$, giving $AB = AC$.

VI

A more intensive study of the bisector problem, from the standpoint of higher plane curves, may afford some deeper insight into the reasons why this innocent-seeming geometrical exercise presents peculiar difficulties. In the analysis is shown the organic relation of the bisector configuration to three well-known curves: the logarithmic spiral, the conchoid, and the cardioid.'

(A) THE LOGARITHMIC SPIRAL

Given the $\triangle ABC$, with AD internal bisector of $\angle BAC$ (Fig. 7). Lay off on CA, $CH = CD$; on AB, $AG = AH$; and on AB produced,

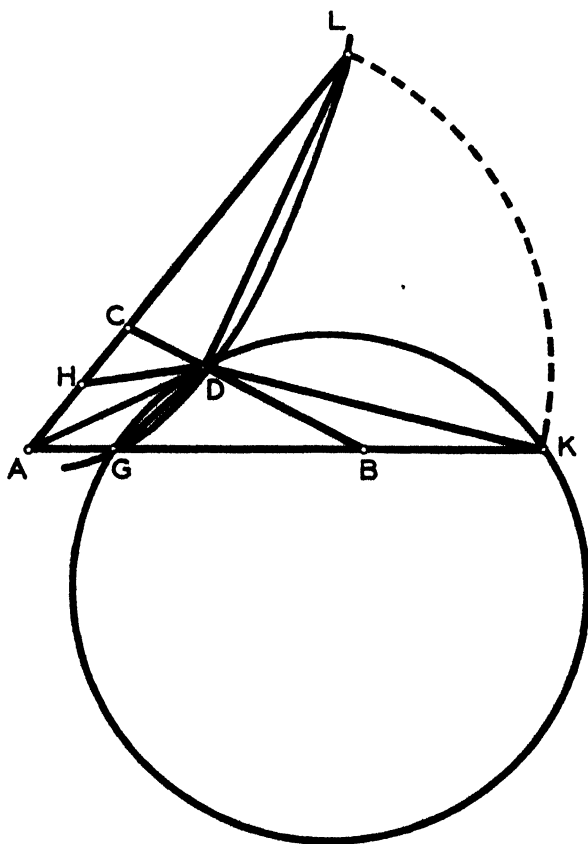


FIG. 7

$BK = BD$. Then describe an arc with D as center, DK as radius, cutting AC produced at L. Join D by straight lines to G, K, and L. Choose $AG = AH = AC - CH = AC - CD$ as unit of measure. Through G and D draw the logarithmic spiral

$$\rho = (AD)^{\frac{2\theta}{\alpha}}$$

where $\angle \alpha = \angle BAC$. If this spiral meets AC produced in L' (not shown in Fig. 7), we have, since $\theta = \alpha$, $\rho = (AD)^2 = AL'$. Then $\triangle AGD$ is similar to $\triangle ADL'$, since $\angle DAG = \angle DAL'$ and $AL':AD::AD:AG (= 1)$.

Now

$$\angle DAH = 180^\circ - \angle AHD - \angle ADH = \angle DAB = 180^\circ - \angle ADB - \angle BDK - \angle BKD$$

giving

$$\angle AHD + \angle ADH = \angle ADB + 2 \angle BKD \dots \dots (1)$$

Moreover

$$\angle CHD + \angle AHD = \angle CDH + \angle ADH + \angle ADB = 180^\circ$$

giving

$$\angle AHD - \angle ADH = \angle ADB \dots \dots \dots (2)$$

From (1) and (2) we have $\angle ADH = \angle BKD$. Consequently $\triangle AHD$ is similar to $\triangle ADK$, and therefore similar to $\triangle ADL$. But since $\triangle AGD = \triangle AHD$, therefore $\triangle AGD$ is similar to $\triangle ADL$. Hence L' falls at L; and we have

$$\overline{AD}^2 = AK = AK \cdot 1 = (AB + BK)(AG) = (AB + BD)(AC - CD)$$

Incidentally we may derive this property by means of the circumcircle of $\triangle GDK$. Since $\angle ADG = \angle ADK$, AD is tangent to this circle at D, giving

$$\overline{AD}^2 = AK \cdot AG = (AB + BK)(AH) = (AB + BD)(AC - CD)$$

(B) THE CONCHOID

The relation of the conchoid to the bisector configuration is even more interesting than that of the logarithmic spiral. Consider the following construction:

(A) CONSTRUCTION. *Through a given point (P) to draw a line, of which the legs of a given angle ($\angle xAy$) cut off a given length l (BC).*

Through the given point P draw a series of lines, laying off the given length l to the right of the vertical side Ax of the given $\angle xAy$, given in position (Fig. 8). Then the locus of the extremities of these lines is a conchoid; and it meets Ay in the required point C, giving the required line PBC. In general, the construction (A) by ruler and compasses alone cannot be made, since it is impossible to satisfy simultaneously

the three conditions: (1) direction of transversal; (2) the falling of B on Ax; (3) the falling of C on Ay. For this purpose a graduated ruler is required.

(B) A SPECIAL CASE. It may happen, however, that the given point P occupy some particular position with reference to the conchoid and $\angle xAy$, which will permit the construction in the special case to be effected by ruler and compasses. This enables us to effect Construction

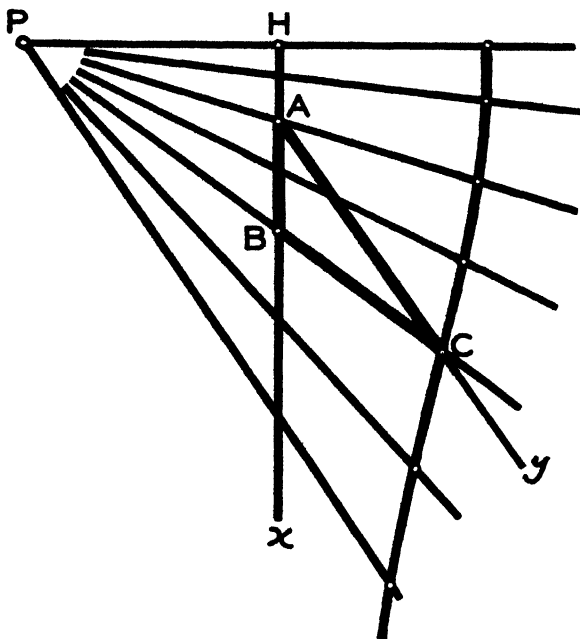


FIG. 8

2, Section IV above: *Given the parts a , A , t_a of a triangle ABC (not drawn), to construct the triangle.*

CONSTRUCTION 4. Take the point P on the bisector Az of the given $\angle xAy$, with $AP = t_a$ (Fig. 9). Erect a perpendicular to AP at P, and lay off, on either side of AP,

$$PQ = PR = a/2 = l/2$$

where l is the given length of Construction (A). Through Q, R draw lines parallel to AP, meeting Ay, Ax in S, T respectively and join S to T. Draw circumcircle of $\triangle AST$, this circle and ST cutting AP produced in

with BC as their common base. From the definition of the cardioid, the same length must be laid off from the circumference on lines drawn through a point in the circumference. Here the circle through A_1, A_2, C, B and cutting perpendicular bisector of BC at M , in N , supplies the circumference with N as the point on it from which the lines for the construction radiate. Let A_1N, A_2N cut BC in K_1, K_2 respectively. Choosing K_2A_2 , the length of the bisector $t_a (= AO, \text{Fig. 2})$, as the given length to be laid off in both directions from the circumference on the rays through N , we construct the cardioid, cutting NK_1A_1 at P . Thus while one extremity of the given length t_a lies on the circle, the other extremity lies on the inner loop of the cardioid, since the bisector of the vertical angle is drawn from the vertex to the base of the triangle. But P , the extremity of $A_1P = t_a = A_2K_2$, must fall on the given base BC (O , extremity of AO , lies on BE and $CF, \text{Fig. 2}$). Since P on the cardioid and K_1 on the line BC must coincide, they both must fall at K_2 . Hence A_1 must fall at A_2 , making $\triangle A_1BC$ congruent to $\triangle A_2BC$. Since this proves that $A_1B = A_2B$, or $AB = AC$ (Fig. 2), the bisector problem is proved.

VII

In this section are submitted three new proofs of the internal-bisector problem, by the indirect method, which are interesting and elegant.

PROOF IX. Given $\triangle ABC$, with AD, BE the equal internal bisectors of the base angles BAC, CBA respectively (Fig. 12). Then we have three cases to consider, depending upon the relative size of the base angles. Suppose, first, $\angle BAC > \angle ABC$. Draw the circumcircle of the triangle AEB , cutting AD produced in Q . Draw a triangle congruent to $\triangle ADB$, with B now at A, A at B , and AD in the new position BD_1 , where D_1 falls on circle with center B and radius BE . Since $\angle BAC > \angle ABC$, AD_1 falls above BE , meeting circumcircle of $\triangle AEB$ in P . Now since $\widehat{PB} < \widehat{EPB}$, chord $PB < \text{chord } EB$. Therefore $BD_1 (= BE) > BP$; and consequently D is outside circumcircle of $\triangle AEB$. Therefore $\angle AD_1B < \angle AEB (= \angle BQA)$. But $\angle AD_1B = \angle BDA$, by construction. Therefore $\angle BDA < \angle BQA$, which is impossible. Hence $\angle BAC \not> \angle ABC$. Second, by similar reasoning, it may be shown that $\angle BAC \not< \angle ABC$. Hence we conclude that $\angle BAC = \angle ABC$, giving $AC = BC$.

PROOF X. Given $\triangle ABC$, with AD, BE the equal internal-bisectors of the base angles BAC, ABC respectively (Fig. 13). Now we have three cases to consider, depending upon the relative size of the base

since C_1B , C_2G are parallel. Hence

$$AC_2 = BC_1 \dots \dots \dots (1)$$

$$\text{and } \angle C_2AD = \angle C_1BE \dots \dots \dots (2)$$

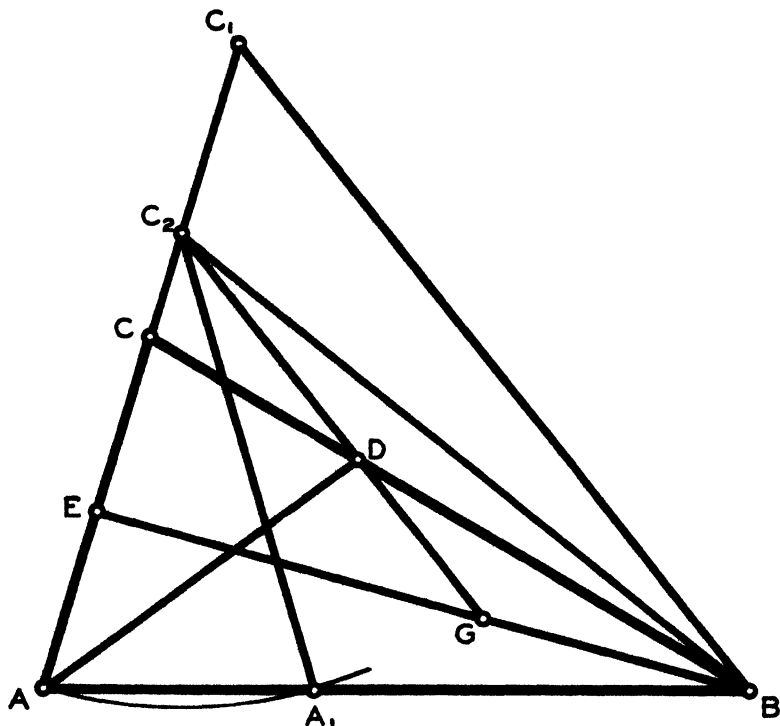


FIG. 13

Now describe arc with center C_2 and radius C_2A cutting AB at A_1 . Since $\angle BAD > \angle ABE$ and $\angle DAC = \angle EBC_1$, we have

$$\angle AA_1C_2 (= \angle BAC) > \angle ABE + \angle EBC_1 (= \angle ABC_1)$$

and *a fortiori*, $> \angle ABC_2$, with A_1 falling between A and B .

Now

$$A_1C_2 = AC_2 = BC_1 \dots \dots \dots (3)$$

by (1) above. But this is impossible; for by joining B to C_2 we have $A_1C_2 < BC_2$ since A_1 falls to the right of the foot of the perpendicular

from C_2 to AB , and $BC_2 < B_1C_1$ since C_1 falls beyond C_2 from the foot of the perpendicular from B to AC_1 , giving $A_1C_2 < BC_1$, which is contradicted by (3). Hence $\angle BAC > \angle ABC$. Similarly, it may be shown that $\angle BAC < \angle ABC$. Hence $\angle BAC = \angle ABC$, giving $AC = BC$.

The third new indirect proof, which follows, like the second, has the merit of employing only theorems of Book I.

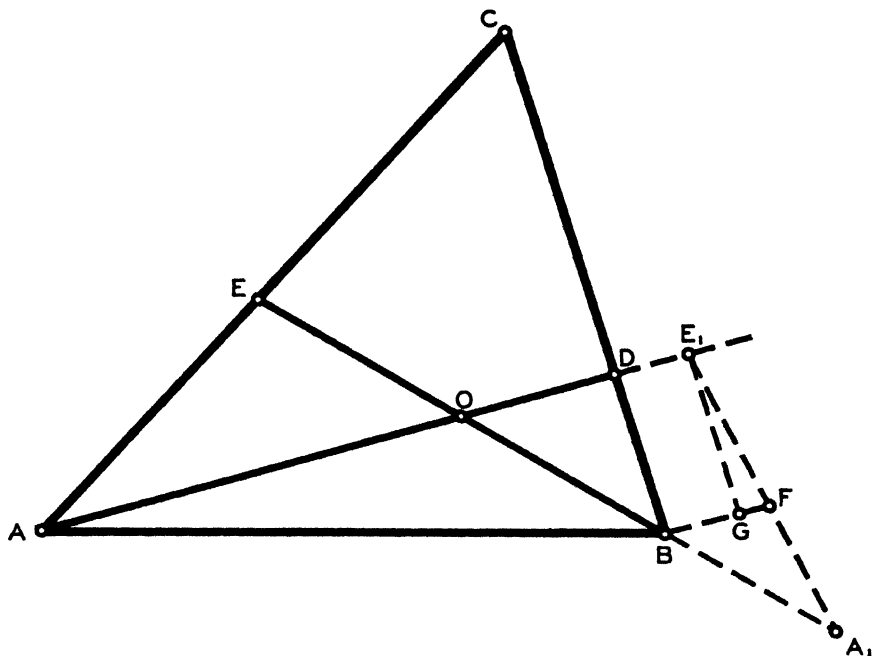


FIG. 14

PROOF XI. Given $\triangle ABC$, with AD , BE , the equal internal-bisectors of the base angles BAC , ABC respectively, intersecting in O (Fig. 14). First let us suppose that $\angle BAC < \angle ABC$. Then $OA > OB$. Since $AD = BE$, therefore $OD < OE$. Extend AD to E_1 , making $OE_1 = OE$, and extend EB to A_1 making $OA_1 = OA$, E_1 lying beyond D from O and A_1 beyond B from O . Join A_1 to E_1 by a straight line.

Now $\triangle OA_1E_1 = \triangle OAE$, giving $\angle OA_1E_1 = \angle OAE$ and $\angle OE_1A_1 = \angle OEA$. Since

$$\angle AOE + \angle OEA + \angle EAO = \angle BOD + \angle ODB + \angle DBO$$

therefore

$$\angle DBO - \angle EAO = \angle OEA - \angle ODB$$

But $\angle OAE < \angle DBO$, by hypothesis, and consequently

$$\angle ODB < \angle OEA \dots \dots \dots (1)$$

Through B draw line parallel to AE_1 , through E_1 a line parallel to DB, these lines meeting at G with G between B and F, since $\angle DE_1G (= \angle ODB) < \angle OE_1A_1 (= \angle OEA)$, from (1). Let BG prolonged meet A_1E_1 at F. Hence

$$DE_1 (= BG) < BF \dots \dots \dots (2)$$

But

$$\angle OEA (= \angle OE_1A_1 = \angle BFA_1) = \angle ACB + \angle EBC$$

Therefore

$$\angle BFA_1 > \angle EBC (= \angle OBD) > \angle BA_1F (= \angle OAE)$$

by hypothesis, giving

$$BA_1 > BF \dots \dots \dots (3)$$

Therefore $BA_1 > DE$, from (2) and (3), or $OA_1 - OB > OE_1 - OD$, giving $OA_1 + OD > OE_1 + OB$ or $AO + OD > OE + OB$ or $AD > BE$, which is contrary to the hypothesis. Hence $\angle BAC < \angle ABC$.

Similarly, with subsidiary figure drawn to left of AC, it may be shown that $\angle BAC > \angle ABC$. Hence $\angle BAC = \angle ABC$, giving $AC = BC$.

VIII

The above investigations, in particular Generalized Theorem 7, Section VI, suffice to disclose the real crux of the problem under discussion. As customarily stated, the internal-bisector problem is misleading, in that it throws the stress upon the wrong features, and veils the essential elements, of the geometrical configuration. The condition that the two equal lines drawn from the *extremities of the base of a triangle* shall be *internal bisectors* of the base angles is really incidental so far as the real problem is concerned. Stress upon the word "triangle" diverts us from the correct mode of approach. The fundamental problem, as we shall see, is that of antiparallels¹² and their relations to the sides of an angle

¹² Definition: If two line-segments B_1C_1 , B_2C_2 , joining points B_1 and C_1 , B_2 and C_2 , of the sides AB, AC respectively of an angle BAC, are such that $\angle AB_1C_1 = \angle AC_2B_2$, the lines B_1C_1 , B_2C_2 are said to be antiparallels with respect to $\angle BAC$.

and its bisector. The essential and indispensable conditions for the generalized theorem are: (1) the lines through a point in the bisector of an angle must meet the sides of the angle; and (2) the segments of the lines, included between the sides of the angle, shall be equal. No triangle is mentioned in the generalized theorem, already proved above, which for convenience will be repeated here:

GENERALIZED THEOREM (7). *If the segments of two lines, drawn through any point in the bisector of an angle and intercepted by its sides, are equal, the bisector is perpendicular to the cross-joins of the corresponding ends of the line segments.*

Immediate and inevitable consequences are that both triangles having the angle-vertex as triangle-vertex and the cross-joins of corresponding ends of the line-segments as bases, are isosceles.

Another theorem intimately connected with this theorem is:

GENERALIZED THEOREM (7a). *Two equal line-segments, included between the sides of an angle and passing through a point in its bisector, are antiparallels.*

Still another theorem of like character is:

GENERALIZED THEOREM (7b). *Two equal line-segments, passing through a point in the bisector of an angle and included between its sides, make equal angles with the bisector.*

This generalized theorem is of fundamental importance, lying as it does on the frontier line between Euclidean and the non-Euclidean geometries. Indeed, it may be said to play a criterial rôle in the theory of parallels. As a substitute for Euclid's Fifth Postulate, upon which Euclidean geometry depends, we may employ **LEGENDRE'S POSTULATE**. *From any point whatever taken within an angle we can always draw a straight line which will cut the two arms of the angle.*¹³

By means of this postulate as a substitute for Euclid's Fifth Postulate, employing the identical figure of the crucial generalized theorem, stated above in three alternative forms, we are enabled to prove Euclid's Fifth Postulate as a theorem, together with the ordinary theory of parallels resulting in the demonstration that the sum of the angles of a triangle make up two right angles.¹⁴ The argument follows.

¹³ Legendre was anticipated in the formulation and use of this hypothesis by J. F. Lorenz: *Grundriss der reinen und angewandten Mathematik* (Helmstedt, 1791).

¹⁴ Adrien Marie Legendre: "Réflexions sur différentes manières de démontrer la théorie des parallèles ou le théorème sur la somme des trois angles du triangle," *Mem. Ac. Sci.* 13: (Paris, 1833). Compare Roberto Bonola: *Non-Euclidean Geometry*, English translation by H. S. Carslaw, pp. 55-60 (Chicago, 1912).

Let Oy and Mu be two straight lines making an acute and a right angle, respectively, with a given line OMz ; and lay off $\angle MOx = \angle MOy$ (Fig. 15). By Legendre's Postulate there passes through M a line meeting the sides of $\angle xOy$ (in A and B). If this line is Mu , the theorem is fulfilled. If this line (AB) is different from Mu , then also a line symmetrical to AB with respect to OMz meets the sides of the $\angle yOx$ (in A' and B'). Then Mu , drawn within the $\angle OMB$, meets Oy (in S), proving for Case I, when one angle is right and the other acute, Euclid's Fifth Postulate for a special case: "Two lines making angles of

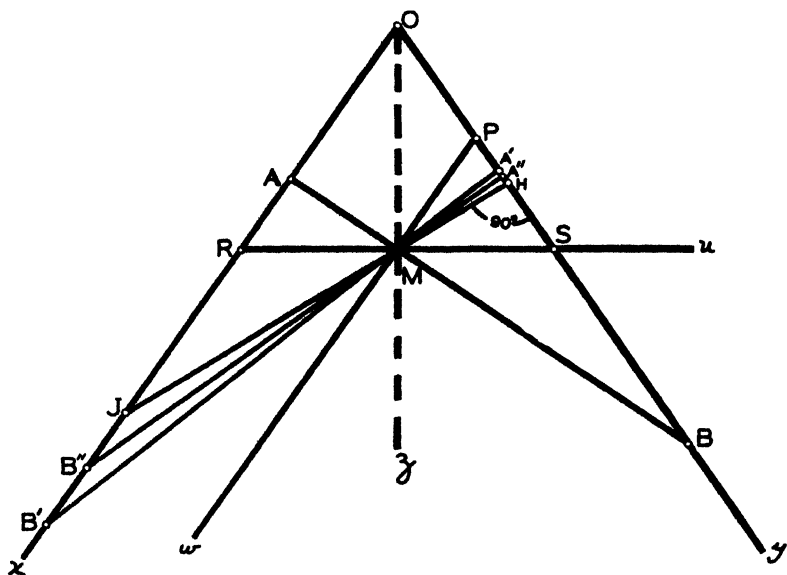


FIG. 15

90° and less than 90° respectively with a given line intersect each other." For Case II, when each of the angles is acute, the theorem is also proved, since any line through M lying within $\angle OMu$ obviously meets Oy .

Let us now consider Case III (Fig. 16). Through M draw a line Mv making with MO an obtuse angle OMv such that $\angle MOy + \angle OMv < 180^\circ$. Lay off $\angle zMt = \angle MOy$, which is acute. Now let fall from M the line MH perpendicular to Oy . If $\angle HMv < 90^\circ$, Mv and Oy meet (Case I). If $\angle HMv = 90^\circ$, vM produced will meet yO produced (Case I). If $\angle HMv = 90^\circ$, we have, since by hypothesis $\angle MOy + \angle Omv < 180^\circ$ and $\angle zMt = \angle MOy$ by construction, Mv

falls within the $\angle OMt$, giving $\angle HMv < 90^\circ$, since Mt is parallel to Oy . Hence Oy and Mv intersect, by Case I, thus proving Euclid's Fifth Postulate for all cases.

We shall next proceed to give two direct demonstrations of the Generalized Theorem (7). It is necessary, in these proofs, to assume the constructions: to draw a parallel, and to draw a perpendicular, to a given line. These constructions, as ordinarily given in the test-books, make use of the circle. If we are to establish our proof by means of the theorems of Book I only, we shall have to show that the two constructions mentioned above may be made by ruler and sect-carrier only, without employing the aid of the circle. Hence we shall set up three lemmas.

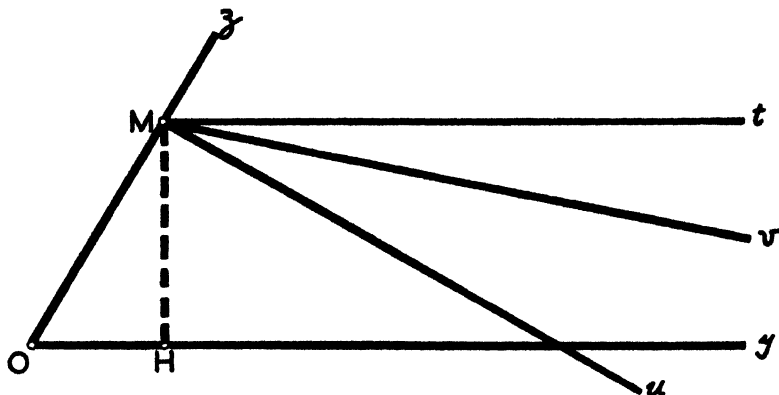


FIG. 16

LEMMA I. *Through a point outside of a line to draw a parallel to the line.* Given line l and point P outside this line. Through P draw a random line to meet line l in Q . Lay off, beyond Q from P , $QR = PQ$. Through R draw another random line to meet l in S , extending RS to T , making $ST = RS$. Then the straight line joining P and T will be parallel to QS .

LEMMA II. *To construct a right angle at a given point.* Through the given point P draw a line at random on which is laid off any convenient length PQ . Through Q draw another line at random, distinct from QP , on which we lay off $QR = QS = QP$. Join R to S and P . Then from the two isosceles triangles PQS , PQR , and the triangle PQS we readily find that the $\angle RPS$ is a right angle.

LEMMA III. *To erect a perpendicular to a given line.* Through P , a

point on the given line l , draw two random lines, distinct from l ; lay off on each any chosen length $PQ = PR$, and from P lay off on l , $PA = PB = PQ$ (Fig. 17). Join A, B to R, Q and let AR meet BQ in S , AQ meet BR in T . Then TS is perpendicular to line l . This follows from Lemma II, giving $\angle AQB = \angle ARB = 90^\circ$, and the theorem in Book I on the concurrence of the three perpendiculars from the vertices of a triangle (ABT) to the opposite sides.

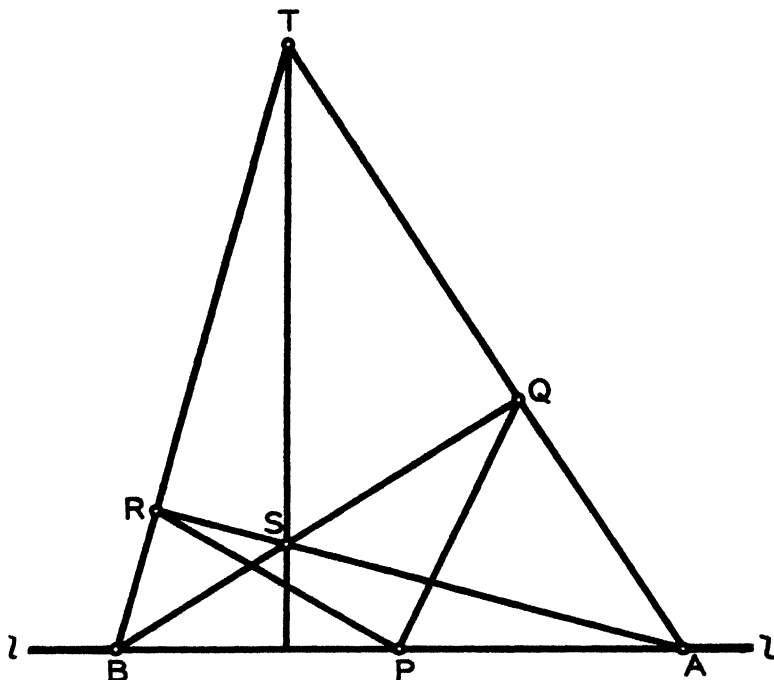


FIG. 17

GENERALIZED THEOREM (7). SECOND PROOF.

Given $\angle xOy$ with bisector Oz , and two equal line segments $AB, A'B'$ drawn through a point M on the bisector Oz (Fig. 15). Through M draw line Pw parallel to Ox (Lemma I), Mu perpendicular to Oz (Lemma III), meeting Oy in S , and the perpendicular to Oy , meeting it in H and Ox in J (Lemmas I and III). Because of symmetry and the nature of parallelism, we need to confine our attention only to lines $A''B''$ drawn within $\angle PMu$. Obviously there are two cases. *Case I.*

We conclude that Oz is perpendicular to AA' and BB' , and hence that the triangles AOA' and BOB' are isosceles. We should note, furthermore, that

$$MA = MA', \quad MB = MB', \quad \angle OMA = \angle OMA', \quad \angle OMB = \angle OMB'$$

Since $\triangle AOB = \triangle A'OB'$, we have also proved Theorem 6, Section IV.

It may be observed that, since $\angle OBM = \angle OB'M$, we may state a theorem intimately connected with Generalized Theorem (7) as follows:

GENERALIZED THEOREM (7c). *If two line-segments, drawn through a point in the bisector of an angle and intercepted by its sides, are equal, they divide proportionally the base angles (both internal and external) of the two triangles, both of them isosceles, formed by the sides of the angle and the cross-joins of the corresponding ends of the two line-segments.*

The following theorems are consequences of the Generalized Theorems (7), (7a), (7b), (7c):

THEOREM 8. *Equal antiparallels to the sides of an angle meet on the bisector of the angle.*

THEOREM 9. *Of two line-segments passing through a point in the bisector of an angle and intercepted by its sides, that one which makes the greater angle with the bisector is the shorter.¹⁵*

As special cases of the generalized theorem we may note the following positions of AB , $A'B'$, in addition to that of internal bisectors of the base angles of $\triangle OBB'$: the medians of the triangle drawn from B , B' to the opposite sides, the symmedians, the perpendiculars to the opposite sides, etc.¹⁶

A straightforward generalization of the internal-bisector problem is the following:

GENERALIZED THEOREM (8). *If two lines, dividing proportionally the*

¹⁵ Theorems (7c), 8 and 9 are variously interconnected.

¹⁶ Concerning the symmedians, consult *L'Intermédiaire des Mathématiciens* 2 (1874), contributions by Alauda, Dellac, Sollertinsky, Lemoine and Tarry. The most elegant indirect proof known, applicable alike to internal bisectors and symmedians, is that of Tarry, *l.c.* Much more elaborate indirect proofs for the internal bisectors are those of Sylvester and B. L. Smith in the former's article, *l.c.* The briefest proof, which is *indirect*, is that referred to in footnote 10. An extended direct proof of the internal bisector problem, by the use of Book I, is that of G. I. Hopkins in E. S. Loomis, *Original Investigation on How to Attack an Exercise in Geometry* (Boston, 1901). Cf. Archibald Henderson, chapter "Mathematics" in *Roads to Knowledge* (New York, 1932). Except for the proof of Hopkins, I know of no direct proofs, by the use of Book I only, except those of the present monograph.

base angles of a triangle internally, are cut by the opposite sides in two equal line-segments, the triangle is isosceles.

A proof, by the indirect method, follows. Given $\triangle ABC$, with Ax , By dividing the base angles $BAC (= \alpha)$, $ABC (= \beta)$ proportionately and meeting the sides AC , BC in D , E respectively, so that $\angle BAD = n\alpha$, $\angle ABE = n\beta$, ($0 < n < 1$), and $AD = BE$ (Fig. 19). Now three

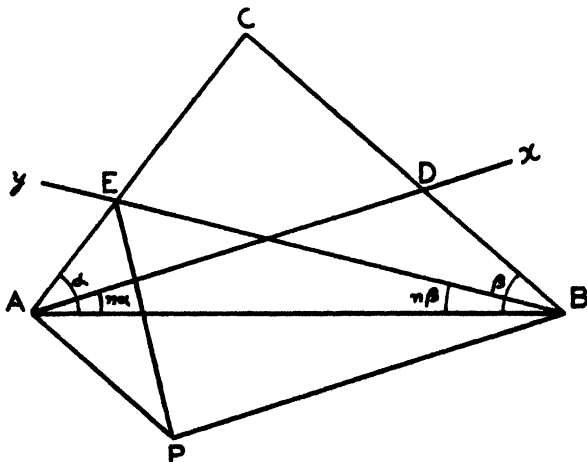


FIG. 19

cases arise, depending upon the relative size of α and β . *Case I.* Suppose $\angle \alpha > \angle \beta$. Then $\angle BAD > \angle ABE$, giving

$$BD > AE \dots \dots \dots (1)$$

Through A , B draw lines parallel to CB , DA respectively, meeting in P , and join P to E . Then $BP = AD = BE$, making

$$\angle BEP = \angle BPE \dots \dots \dots (2)$$

But since $PA (= BD) > AE$, from (1),

$$\angle AEP > \angle APE \dots \dots \dots (3)$$

Adding (2) and (3) we have

$$\angle BEA > \angle BPA \dots \dots \dots (4)$$

Now

$$\angle BAE + \angle ABE + \angle BEA = \angle ABP + \angle BAP + \angle BPA = 180^\circ$$

or

$$\angle \alpha + \angle n\beta + \angle BAE = \angle n\alpha + \angle \beta + \angle BPA$$

Hence

$$\angle BAE - \angle BPA = (n - 1) \angle (\alpha - \beta)$$

which is impossible, since the left side of the equation and $\alpha - \beta$ are positive, but $n - 1$ is negative. *Case II.* If we assume $\angle \alpha < \angle \beta$, a similar contradiction may be shown, by drawing through A, B lines parallel to EB, CA respectively. Hence $\angle \alpha = \angle \beta$, and therefore $AC = BC$.¹⁷

By the use of the formula for the length of the internal bisector, we find by setting $\overline{AD}^2 = \overline{BE}^2$,

$$c(a - b)(a + b + c)(c^3 + 3abc + bc^2 + ab^2 + ac^2 + a^2b) = 0$$

giving the unique solution $a = b$. If however we adopt the same method for the external bisectors of the same base angles, a different situation develops, since we find

$$c(a - b)(a + b + c)(c^3 + 3abc - bc^2 - ab^2 - ac^2 - a^2b) = 0$$

giving two possible solutions: $a = b$ (1) and $c^3 + 3abc - bc^2 - ab^2 - ac^2 - a^2b = 0$ (2). Equation (1) gives the isosceles triangle case; equation (2) may be put in the form $4Rr_o = ab + c^2$, where R is the radius of the circumcircle of the triangle and r_o is the radius of the circle externally tangent to the side AB.¹⁸

GENERALIZED THEOREM (7). THIRD PROOF. Given $\angle A$ with bisector AO, and equal line segments BOE, COF intercepted between sides of $\angle A$ (Fig. 20). Drop a perpendicular on AO produced, meeting it at N and EB at L (Lemmas II and III). Extend LN to L_1 , making $NL_1 = LN$. Draw line through O and L_1 , meeting the sides AB,

¹⁷ Of this theorem I have seen no direct proof.

¹⁸ *Mathesis* 15: 261-2 (1895). Compare also Sylvester, *l.c.* for a trigonometric analysis which merely skirts the edges of the problem of the external bisectors. Apparently no solution, solely by geometrical methods as expressed by the condition $4Rr_o = ab + c^2$, is available. Consult, however, W. E. Heal: Paper II. Relating to the Demonstration of a Geometrical Theorem, *American Mathematical Monthly* 25: 182-3 (1918).

AC of the given angle BAC at E_1, B_1 respectively. Since $\angle NOE_1 = \angle NOE$ and $\angle EOB_1 = \angle EOB$, therefore $\angle NOE_1 = \angle NOE$ and

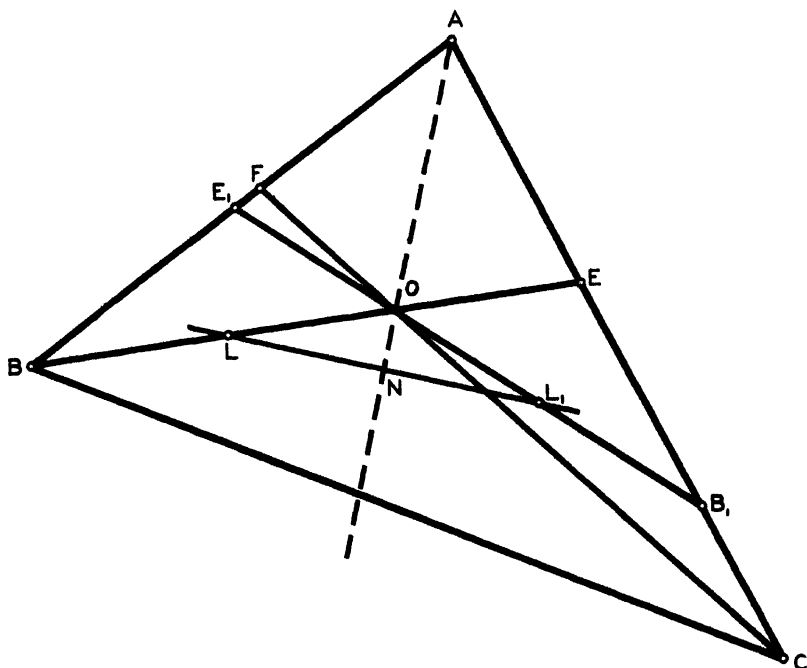


FIG. 20

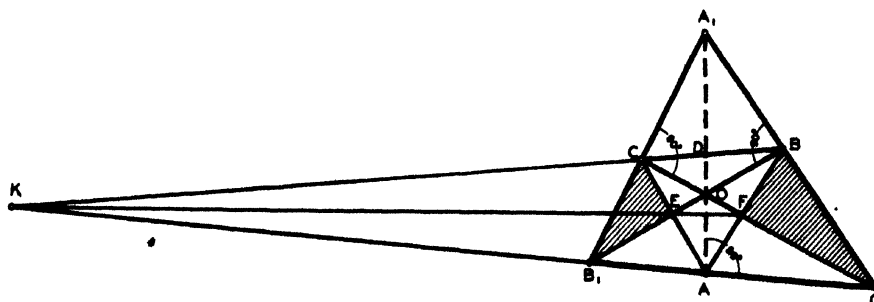


FIG. 21

$\angle AOE_1 = \angle AOE$. Hence $\triangle AOE_1 = \triangle AOE$, giving $AE_1 = AE$ and $\angle AE_1O = \angle AEO$. There $\triangle A_1E_1B_1 = \triangle AEB$. Therefore $AB_1 = AB$

and $E_1B_1 = EB$. From the symmetry of the figure about AO , the continuous increase of length of intercepted segment from RS to Pw (Fig. 15) and the uniqueness of the construction of E_1B_1 , it follows that FC must coincide with E_1B_1 . Hence $EB = E_1B_1 = FC$; and therefore $AB = AC$, and AO is perpendicular to EF and BC at their respective mid-points.

IX

Since the lines AB, AC, BE, CF constitute the nucleus of a complete quadrilateral, the generalized theorem falls in the domain of projective geometry. It nevertheless has a semi-metrical character, because of the condition that the segments of the lines through a point on the bisector of the given angle and intercepted between its sides, are equal. The following proof of the internal-bisector problem brings into clear relationship the projective, harmonic, and metric features of the theorem,

Given $\triangle ABC$, with internal bisectors AD, BE, CF of the angles A, B, C , meeting at O (Fig. 21). Prolong BE and CF to meet the perpendicular to AO at A , in the points B_1, C_1 respectively. Join B_1 to C and C_1 to B . Since AO and AB_1 are the internal and external bisectors of $\angle BAC$, therefore $BOEB_1$ and $COFC_1$ are harmonic ranges with O as self-corresponding point. Therefore BC, FE, C_1B_1 are concurrent in the point K ; and since the triangles BFC_1, CEB_1 are in perspective, then by Desargues's Theorem, $B_1C, C_1B; B_1E, C_1F; CE, BF$ meet in the collinear points A_1, O, A respectively. Now BC and FE are perpendicular to AOA_1 [Generalized Theorem (7) Section VIII], and hence parallel to C_1B_1 . Hence K goes to infinity; and D , paired with K in the harmonic range $BDCK$, bisects BC . Accordingly $AB = AC$.

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NOTES ON THE GENUS *FLINDERSIA* R. Br. AND THE SYSTEMATIC ANATOMY OF THE IMPORTANT FLINDERSIAN TIMBERS INDIGENEOUS TO QUEENSLAND

By ELLWOOD S. HARRAR

PLATES 24-26

INTRODUCTION

The genus *Flindersia* R. Br., since it was first recognized as distinct from other related groups, has been subject to considerable taxonomic disagreement among systematists. Bentham and Hooker (1862-1867) originally placed *Flindersia* in the Meliaceae. Bailey (1899-1902) likewise classified it with this family when he compiled "The Queensland Flora." Engler and Gilg (1924) noted the occurrence of secretory tissue in the leaves and cortex of several species and promptly transferred the genus to the Rutaceae, although Hutchinson (1926) still holds to the concepts of Bentham and Hooker. Welch (1931) suggests that the group might occupy a position midway between the Rutaceae and the Meliaceae, and White (1931), after carefully examining floral characters, states that *Flindersia* appears to be more closely related to the Rutaceae than to the Meliaceae but favors the formation of a new family, Flindersiaceae.

The present study has a two-fold purpose: (1) to present information of diagnostic value concerning the important commercial timbers of this genus which are indigenous to Queensland, and (2), to ascertain if there is sufficient evidence, based upon wood anatomy, to determine accurately the correct taxonomic position of *Flindersia*.

SIZE AND DISTRIBUTION

According to White (1921) *Flindersia* comprises 18 species of trees only 3 of which occur naturally beyond Australian shores. The Queensland flora includes 14 or 15 species; these are widely scattered throughout the state, some occurring in the rain-forests and others in much less humid regions. Economically the group is rather important and is

especially noted for the very fine cabinet woods and structural timbers which it produces.

MATERIALS

The materials which served as a basis for this study were obtained from the Queensland Service through the coöperation of the Provisional Forestry Board, Brisbane, Queensland. Herbarium sheets from the identical tree accompanied each specimen of wood and all were checked for accuracy of identification by government botanists before shipment to the United States. The wood samples now reside in the collections of the Department of Wood Technology, The New York State College of Forestry at Syracuse, New York, and the herbarium material is on file at the New York Botanical Garden, Bronx Park, New York City. Additional wood samples, many of them trade samples from the collections of the New York State College of Forestry, and from those of the College of Forestry, University of Washington, Seattle, Washington, were used for purposes of comparison and for the collection of additional anatomical data. The species examined are indicated below:

1. *F. acuminata* C. T. W.
2. *F. australis* R. Br.
3. *F. bennettiana* F. v. M.
4. *F. bourjotiana* F. v. M.
5. *F. brayleyana* F. v. M.
6. *F. collina* Bail.
7. *F. ifflaiana* F. v. M.
8. *F. oxleyana* F. v. M.
9. *F. pubescens* Bail.
10. *F. schottiana* F. v. M.

CHARACTERISTICS OF FLINDERSIAN WOOD

Physical properties. Sapwood creamy white to silvery white or yellowish white, gradually merging into somewhat darker heartwood, except in those woods where these two zones cannot be differentiated; heartwood creamy white, silvery white, pale yellow, light brownish yellow, light golden brown or rarely mahogany-red or coppery red; dull or with a pronounced pearly lustre or silky sheen; occasionally oily to the touch; with an aromatic odor, rancid, or odorless; in weight ranging from approximately 35 to 60 lbs. per cubic foot air-dry; usually even-

textured; straight, undulate, or firmly interlock-grained; plainly or handsomely figured.

Anatomical features

Growth rings occasionally evident at high magnifications but not sharply defined, usually delineated by narrow to relatively wide lines or bands of dense fibrous tissue where growth increments adjoin.

Vessels variable in size but of nearly uniform diameter within a species, oval, orbicular, and polygonal (x),¹ solitary, in 2-8 (mostly 2-4) radial rows, and occasionally in small clusters not exceeding 6, usually surrounded by a narrow sheath of parenchyma which is often interrupted by fibers and wood rays flanking the vessels; vessel members variable in length, truncate or with short tails at one or both ends, commonly plugged at the junctures or wholly occluded with yellow to reddish brown gummy infiltration or white granular deposits; perforation plates simple, nearly horizontal; inter-vessel pits irregularly polygonal, crowded, 4-8 microns in width; pits leading from vessels to ray and longitudinal parenchyma similar to but smaller than those of the inter-vessel type; tyloses wanting.

Fibers usually less than 30 microns in diameter, thin- to extremely thick-walled, non-septate, aligned in radial rows which are further aggregated into extensive tracts; inter-fiber pits minute, orbicular to slit-like, bordered.²

Longitudinal parenchyma metatracheal, paratracheal, and diffuse, in cambiform rows of 2-8 units along the grain, occasionally further divided into crystal loculi: (a) *metatracheal* parenchyma abundant (in certain woods this is the most conspicuous feature on the transverse section), the cells forming 2-40 plus (mostly 2-12) seriate bands (x) which occur with considerable regularity in some woods and very irregularly in others; (b) *paratracheal* parenchyma usually forming a 1-3 seriate sheath, often dissected by fibrous tissue and rays; (c) *diffuse* parenchyma seldom conspicuous, the cells occurring as isolated individuals scattered through the tracts of fibrous tissue; cells of all types occasionally crystalliferous, (except in *F. bourjotiana*) those of the "a" and "b" types occasionally thick-walled; inter-parenchymatous pits minute.

Wood rays plainly visible or indistinct without a lens, unstoried,

¹ x. r. t. = transverse, radial, and tangential surfaces respectively.

² Pit borders in the very thick-walled fibers are usually poorly developed and in some instances appear to be wholly lacking.

homogeneous, or rarely bordering on the heterogeneous type, heterogeneous in *F. collina*, 1-7 seriate, of nearly uniform width within a species, but quite variable in height; inter-ray cell pits minute; gummy infiltration sparse; crystals wanting (except in the marginal upright ray cells in *F. collina*).

Gum canals present at wide intervals, restricted to the broader bands of metatracheal parenchyma, solitary or in tangential rows of 2-many.

Pertinent features of the species

This study has revealed that flindersian timbers possess numerous physical and anatomical departures of sufficient magnitude to permit of their specific identification. To facilitate comparisons, the significant properties of each have been noted and recorded in Table I. A key for these woods, based on both their microscopic and macroscopic characteristics, also has been compiled.

KEY TO THE WOODS

1. Heartwood reddish brown, mahogany-red, or copper-colored, aromatic
F. brayleyana
1. Heartwood ranging through shades of creamy white, silvery white and grayish white to yellow-brown, golden brown or brownish white, never tinged with red, odorless or occasionally rancid, but never aromatic..... 2
2. Heartwood with a rancid odor, oily to the touch..... 3
2. Heartwood odorless or if scented not rancid, not oily to the touch..... 4
3. Wood rays plainly visible without a lens, 1-6 seriate; ray cells 40-50 microns in diameter (t); metatracheal parenchyma cells thin-walled..... *F. australis*
3. Wood rays not discernible without a lens, 1-3 seriate; ray cells 30-35 microns in diameter (t); metatracheal parenchyma cells thick-walled..... *F. iflaiana*
4. Vessel orifices plainly visible without a lens..... 5
4. Vessel orifices indistinct without a lens..... 7
5. Fiber walls uniformly thin; metatracheal parenchyma very sparse
F. acuminata
5. Fiber walls variable in thickness, those in the outer limits of growth rings the thickest..... 6
6. Wood rays 1-5 seriate, 32-40 cells high; growth rings poorly defined; longitudinal parenchyma commonly crystalliferous..... *F. pubescens*
6. Wood rays 1-3 seriate, 24-30 cells high; growth rings sharply defined; longitudinal parenchyma nearly or wholly devoid of crystals... *F. bourjotiana*
7. Wood rays 1-2 seriate, 12-18 cells high; growth rings sharply defined.
F. schottiana
7. Wood rays 1-7 seriate, 30-52 cells high; growth rings poorly defined..... 8
8. Vessels mostly in 2-4 radial rows (x), 14-28 per mm². metatracheal parenchyma sparse..... *F. ozleyana*
8. Vessels mostly solitary and radially paired (x), 10-17 per mm²; metatracheal parenchyma abundant..... 9

TABLE I
Diagnostic features of the woods

| SPECIES | HEARTWOOD | | | | SEASONAL RINGS | VENEERS | | | | | |
|-----------------------|---|----------|------|---|-----------------|---------------------------------|----------------------------|---|------------------------------|----------------------------------|-------------|
| | Color | Odor | Feel | Average weight in pounds per cubic foot | | Orifices visible without a lens | Arrangement | Minimum and maximum No. per mm ² | Average and maximum diameter | Average length of vessel members | |
| <i>F. acuminata</i> | Creamy white to light silvery brown, lustrous | None | Dry | 35 | Poorly defined | Yes | Mostly paired | | 5-15 | 200-250 microns | 600 microns |
| <i>F. australis</i> | Yellowish brown to golden brown, dull | Rancid | Oily | 50 | Not evident | Yes | Mostly paired | | 12-20 | Same | 650 microns |
| <i>F. bennettiana</i> | Brownish white to pale straw-colored, lustrous | None | Dry | 55 | Poorly defined | No | Mostly paired | | 10-15 | 150-175 microns | 450 microns |
| <i>F. bourjotiana</i> | Creamy white to brownish white, lustrous | None | Dry | 45 | Sharply defined | Yes | Mostly solitary and paired | | 7-15 | 175-210 microns | 650 microns |
| <i>F. brayleyana</i> | Reddish brown to mahogany-red or copper-colored | Aromatic | Dry | 40 | Sharply defined | Yes | Same | | 5-15 | 175-225 microns | Same |
| <i>F. collina</i> | Creamy white, lustrous | None | Dry | 60 | Poorly defined | No | Same | | 10-17 | 130-175 microns | 450 microns |
| <i>F. iffiana</i> | Golden yellow to yellowish brown, somewhat lustrous | Rancid | Oily | 60 | Not evident | Yes | Mostly in 3-6 radial rows | | 8-20 | 200-280 microns | 700 microns |
| <i>F. ozleyana</i> | Light golden brown to yellowish brown, lustrous | None | Dry | 50 | Poorly defined | No | Mostly in 2-4 radial rows | | 14-28 | 110-150 microns | 525 microns |
| <i>F. pubescens</i> | Silvery white to light silvery brown, lustrous | None | Dry | 40 | Poorly defined | Yes | Mostly paired | | 6-9 | 200-240 microns | 625 microns |
| <i>F. schottiana</i> | Silvery gray to brownish white, lustrous | None | Dry | 45 | Sharply defined | No | Same | | 12-20 | 125-175 microns | 450 microns |

| SPECIES | FIBRES | | LONGITUDINAL PARENCHYMA* | | | | RAYS | | | | |
|-----------------------|--|---------------------------------|--|---|------------------------|-----------------|--|--|--------------------|------------------------------|--|
| | Walls | Pits | Metatracheal | Paratracheal | Visible without a lens | Series- tion | Ave. and max. height in cells | No. per mm. across the grain | Composi- tion | Ave. dia. of ray cells | |
| <i>F. acuminata</i> | Uniformly thin | Orbicular | Sparse, thin-walled, in 1-6 seriate bands | 1-3 (mostly 1) seriate | Yes | 1-5 | 32-41 | 4-6 | Homo- geneous | 16-24 microns | |
| <i>F. australis</i> | Uniformly very thick | Slit-like | Abundant, thin-walled, in 1-8 seriate bands | 1-2 seriate | Yes | 1-6 | 42-54 | 3-5 | Homo- geneous | 40-50 microns | |
| <i>F. bennettiana</i> | Variable, thickest in outer limits of ring | Lenticular to slit- like | Abundant, thin-walled, in 1-7 seriate bands | Uniseriate | Barely | 1-6 | 40-52 | 3-6 | Homo- geneous | 15-20 microns | |
| <i>F. bourjoiana</i> | Same | Same | Abundant, thin-walled, in 1-7 seriate bands | 1-2 seriate | Barely | 1-3 | 18-33 | 4-7 | Homo- geneous | 25-30 microns | |
| <i>F. brayleyana</i> | Same | Lenticular | Abundant, thin-walled, in 1-40 (mostly 1-12) seriate bands | Sparse, occur- ring as an occasional cell or in 2's and 3's | Barely | 1-4 | 24-30 | 3-5 | Homo- geneous | 18-22 microns | |
| <i>F. collina</i> | Same | Slit-like | Abundant, thin-walled, in 2-12 seriate bands | 1-2 seriate | Yes | 1-7 | 31-51 | 4-6 | Hetero- geneous | 25-30 microns | |
| <i>F. iffaiana</i> | Uniformly thick | Same | Abundant, thick-walled, in 1-7 seriate bands | Uniseriate | No | 1-3 | 25-36 | 6-9 | Homo- geneous | 30-35 microns | |
| <i>F. ozleyana</i> | Variable, thickest in outer limits of ring | Orbicular to len- ticular | Sparse, thin-walled, in 2-10 seriate bands | 1-4 seriate | Yes | 1-6 | 34-48 | 4-6 | Homo- geneous | 25-30 microns | |
| <i>F. pubescens</i> | Same | Same | Abundant, thin-walled, in 2-10 seriate bands | 1-3 seriate | Yes | 1-5 | 32-40 | 4-7 | Homo- geneous | 25-35 microns | |
| <i>F. schottiana</i> | Same | Same | Abundant, thin-walled, in 2-7 seriate bands | 1-2 seriate | No | 1-2 | 12-18 | 3-6 | Homo- geneous | 25-30 microns | |

* Crystalliferous cells abundant except in *F. bourjoiana*.

9. Wood rays heterogeneous, the cells 25-30 microns in diameter (t), occasionally crystalliferous; metatracheal parenchyma bands 2-12 seriate.... *F. collina*
9. Wood rays homogeneous, the cells 15-20 microns in diameter (t), non-crystalliferous; metatracheal bands 1-7 seriate *F. bennettiana*

DISCUSSION

Since the pertinent physical and anatomical properties which characterize flindersian wood have been already reviewed, it now becomes expedient to compare them with those which feature the Meliaceae and Rutaceae, respectively.

The timbers of the Meliaceae, as well as those of the Rutaceae, themselves exhibit a wide range of physical and anatomical properties. On the other hand, there are several species included in each of these two families which possess numerous features in common. Woods of this sort not infrequently suggest close botanical affinity, and in many instances it is only after they have been critically examined that their real taxonomic relationships become apparent. Included among woods of this type are those produced by members of the genus *Flindersia*. In this group of woods the arrangement of vessels and the distribution of longitudinal parenchyma closely resembles that of many rutaceous, as well as a large number of meliaceous woods. The perforation plates, inter-vessel pitting, pits between vessels and ray or longitudinal parenchyma are likewise all so similar that they are of practically no value diagnostically. It is, therefore, rather obvious that the true systematic position of *Flindersia* can be determined only after comparisons have been made with those physical and anatomical features which alone give real character respectively to the Meliaceae and to the Rutaceae.

Meliaceous woods are pink, red, or reddish brown for the most part, while those belonging to the Rutaceae usually vary through shades of lemon-yellow to light yellowish brown or brownish white. With respect to color the heartwood of *F. brayleyana* is typical of many woods belonging to the mahogany family,³ but the colors of the nine other species examined are much more characteristic of woods included in the rue family. Scented woods are featured by several genera of both families. Those included in the Meliaceae are commonly aromatic, while in the Rutaceae ill-scented ones are occasionally found. The timbers of *F. australis* and *F. ifflaiana* are noticeably rancid, while those of *F. brayleyana* and *F. laevicarpa* possess pleasant aromas.

³ Swain (1928) reports that the timbers of *F. pimentiliana* F. v. M. and *F. laevicarpa* C. T. W. possess a reddish hue.

An element of nearly universal occurrence in meliaceous wood is the septate fiber. According to Kribs (1930), fibers of this sort are developed in all members of the large sub-family, Swietenioideae, as well as in the vast majority of those woods included in the sub-family Melioideae. They are wanting, however, in the single genus *Lovoa* of the sub-family Lovoinoideae. Septate fibers have never been observed in woods comprising the Rutaceae, nor do they occur in flindersian timbers.

Structural differences in the wood rays are also diagnostic and offer additional means for differentiating between many woods of these two families. In the Meliaceae, the heterogeneous ray, like the septate fiber is a nearly constant feature of the wood. Rays of this sort occur in all of the woods in the Swietenioideae (Kribs, 1930), and in most of the Melioideae. In fact only four genera belonging to the mahogany family, namely, *Lovoa*, *Ekebergia*, *Walsura*, and *Azadirachta* include woods which possess neither heterogeneous rays nor septate fibers. Where heterogeneity exists, two types of rays are commonly present. The larger and more conspicuous of the two is usually multiseriate and is composed of procumbent cells through the medial portion, and then is tapered above and below to uniseriate margins consisting of one to several rows of upright cells. In woods belonging to the Swietenioideae the marginal upright cells commonly contain crystals. The smaller ray, by contrast, is ordinarily uniseriate and consists entirely of upright cells.

Narrow homogeneous rays feature woods belonging to the Rutaceae, although in several species the marginal ray cells are greatly restricted in length radially across the grain thus resulting in a structure bordering on the heterogeneous type. Marginal cells of this sort are devoid of crystals.

Ripple marks traceable to storied wood rays feature several genera of the Meliaceae. In rutaceous wood they have been observed only in members of the genus *Chloroxylon*. The taxonomic position of this genus, however, is debatable and its inclusion in this family appears to be more or less provisional.

Except in *F. collina* the rays of all flindersian woods examined are homogeneous or merely border on the heterogeneous type in the manner described above, and neither the marginal nor body ray cells contain crystals. The rays of *F. collina*, on the other hand, are strongly heterogeneous, and many of the marginal upright cells are crystalliferous, features exhibited in woods belonging to Swietenioideae of the Meliaceae. None of the ten woods examined has storied wood rays.

Gum canals have been observed in many genera of both families,

hence their occurrence in *Flindersia* is of little diagnostic significance here.

Based upon modern taxonomic concepts *Flindersia* appears to be more closely related to the Rutaceae than to the Meliaceae. From the discussion just completed it is evident that a nearly parallel situation exists when viewed from the standpoint of wood anatomy. Neither the taxonomic nor anatomical characteristics point clearly to either of these two families, but rather appear to combine certain features of both. In conclusion the writer feels that the anatomical evidence gathered during this study lends further support to White's (loc. cit.) views; namely that *Flindersia* should be set up as a separate family, the Flindersiaceae.

SUMMARY

1. In Queensland the timbers produced by the genus *Flindersia* are commonly sold in mixture and thus lose their identity in the trade. In recent years several of them have been used in the United States for cabinet purposes. A systematic study of the wood from 10 of the more important species resulted in obtaining numerous physical and anatomical departures of sufficient magnitude to permit of specific identification.

2. Flindersian wood exhibits physical and anatomical features which are neither wholly rutaceous nor meliaceous, but rather combine several characteristics of the rue and mahogany families, respectively.

3. The formation of a new monotypic family, Flindersiaceae, to receive the genus *Flindersia* is favored by the writer.

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BIBLIOGRAPHY

- BAILEY, F. M., 1899-1902. The Queensland Flora. 1: 181.
BENTHAM AND HOOKER, 1862-1867. Genera Plantarum. 1: 340.
ENGELER AND GILG, 1924. Syllabus der Pflanzenfamilien. p. 248.
HUTCHINSON, J., 1926. The Families of Flowering Plants.
KRIBS, D. A., 1930. Comparative Anatomy of the Woods of the Meliaceae. Am. Jour. Bot. 17: 724-738.
SWAIN, E. H. F., 1928. The Timbers and Forest Products of Queensland. A. J. Cummings, Government Printer, Brisbane.
WELCH, M. B., 1931. The occurrence of Intercellular Canals in the Wood of Some Species of *Flindersia*. Jour. Proc. Roy. Soc. N. S. W. 64: 362.
WHITE, C. T., 1921. Notes on the Genus *Flindersia*. Proc. Linn. Soc. N. S. W. 46: 324.
———, 1931. Tropical Woods. No. 25. p. 18, Footnote.

EXPLANATION OF PLATES

PLATE 24

- Fig. 1. Transverse section of *Flindersia acuminata*, $\times 35$.
Fig. 2. Tangential section of *Flindersia acuminata*, $\times 35$.
Fig. 3. Transverse section of *Flindersia australis*, $\times 35$.
Fig. 4. Tangential section of *Flindersia australis*, $\times 35$.
Fig. 5. Transverse section of *Flindersia bennettiana*, $\times 35$.
Fig. 6. Tangential section of *Flindersia bennettiana*, $\times 35$.

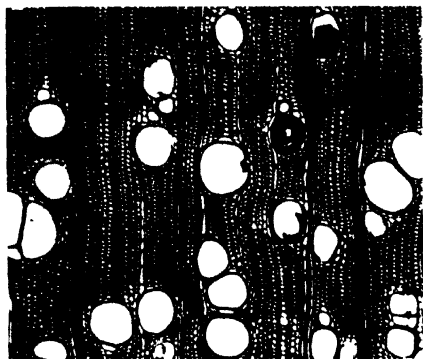
PLATE 25

- Fig. 1. Transverse section of *Flindersia bourjotiana*, $\times 35$.
Fig. 2. Tangential section of *Flindersia bourjotiana*, $\times 35$.
Fig. 3. Transverse section of *Flindersia brayleyana*, $\times 35$.
Fig. 4. Tangential section of *Flindersia brayleyana*, $\times 35$.
Fig. 5. Transverse section of *Flindersia collina*, $\times 35$.
Fig. 6. Tangential section of *Flindersia collina*, $\times 35$.

PLATE 26

- Fig. 1. Transverse section of *Flindersia iflaiana*, $\times 35$.
Fig. 2. Transverse section of *Flindersia pubescens*, $\times 35$.
Fig. 3. Transverse section of *Flindersia ozleyana*, $\times 35$.
Fig. 4. Tangential section of *Flindersia ozleyana*, $\times 35$.
Fig. 5. Transverse section of *Flindersia schottiana*, $\times 35$.
Fig. 6. Tangential section of *Flindersia schottiana*, $\times 35$.

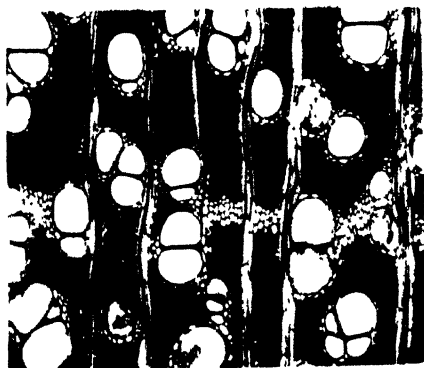
PLATE 24



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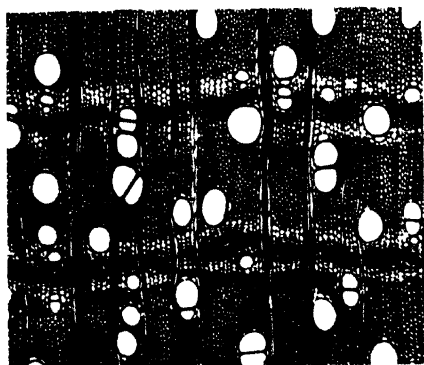
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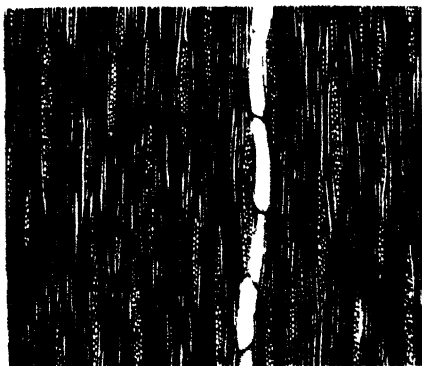
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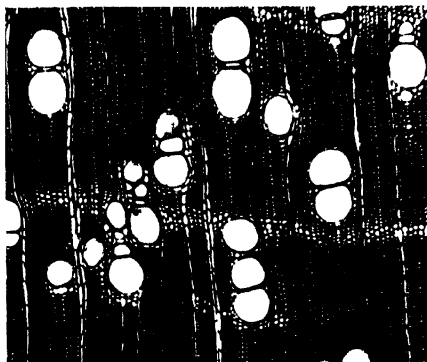


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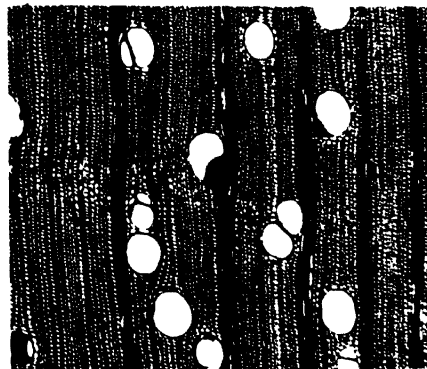
PLATE 25



1.



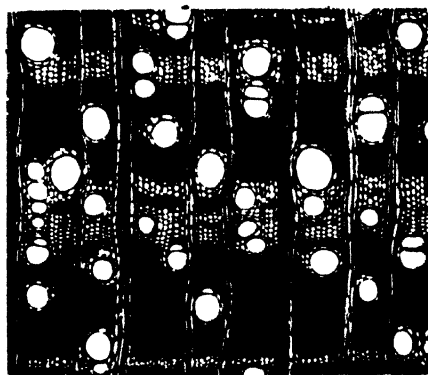
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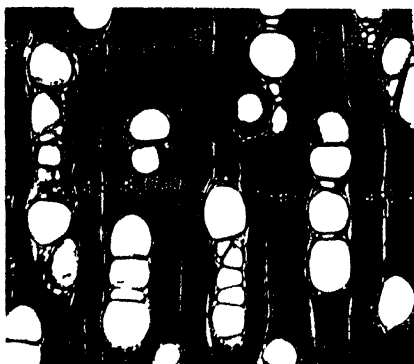


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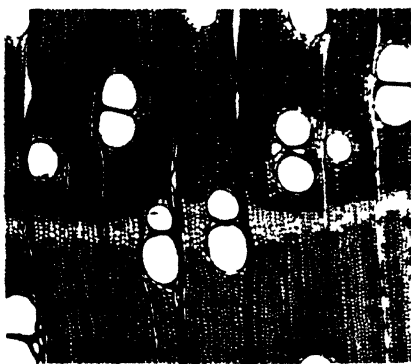


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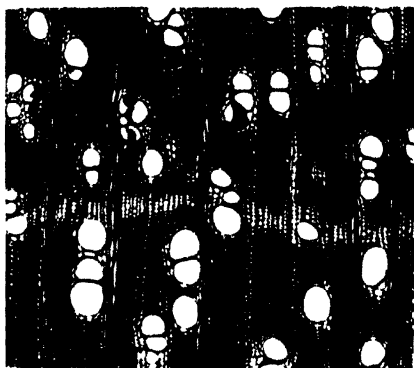
PLATE 26



1.



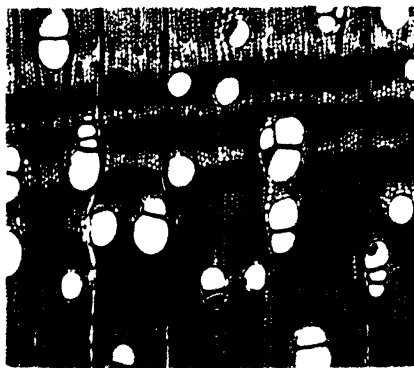
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5.



6.

THE DESTRUCTIVE DISTILLATION OF PEANUT HULLS

By WALKER F. HUNTER, JR.¹ and R. W. BOST

INTRODUCTION

Numerous laboratories have attempted to develop processes for the utilization of bulk farm waste products in this country with little success. Of these waste products, perhaps the most logical to utilize are the so-called pentosan waste products, since they are already concentrated in industry. The most outstanding utilization so far has been the development of furfural and furan derivatives by the Quaker Oats Company.

For the work described in this paper peanut hulls were used, although most of the material may apply equally as well to the other products, such as wheat and other grain hulls, corn cobs, and certain straw and stalks of farm waste. Over 70,000 tons of peanut hulls are collected yearly in southern shelling and processing plants. When all these farm products are considered, the waste staggers the imagination. It is true that these products have found many uses to date, but they are always a burden and not an asset to the concern that has them. For fuel they are locally used at the site of production, but due to their bulk, transportation is out of the question. Otherwise, they are supplied to a small local market free or at a minimum cost as substitutes for other cheap products.

Certain researches have been carried out on peanut hulls with little success and no commercial advancement. Though they contain upward of 40% cellulose, it has been found by the U. S. Farm Waste Division to be an uneconomical source of alpha cellulose.² This result is due to low yield of alpha cellulose and the drastic and expensive processing that must be applied to free the cellulose from the other constituents of the hulls.³ Though peanut hulls might lend themselves to the production of furfural as well as oat hulls, no one has seen fit to compete commercially with this source.^{4,5} Fermentation, both anaerobic and

¹ Present address, Eastman Kodak Co., Rochester, N. Y.

² Anon., Chem. and Met. Eng., **37**: 417 (1930).

³ Lynch and Goss, Ind. and Eng. Chem., **23**: 903 (1930).

⁴ Hall, Slater, and Acree, Bur. Standards J. Research, **4**: 329 (1930).

⁵ Bryner, Christensen and Fulmer, Ind. and Eng. Chem., **28**: 206 (1936).

aerobic, has been investigated but low yields were obtained, due, it is believed, to the inhibiting action of the lignin present.⁶

It appears, today, that wood distillation from a commercial point of view is rapidly losing its prestige. The economic condition of this industry is largely due to competition from synthetic chemicals and also to the increased cost of purifying the products distilled from wood. If the cost of raw material could be lowered and technical advance could be made in the industry, destructive distillation might obtain a better footing economically. This research was instigated to determine the relationship between the products of distillation from pentosan waste products such as peanut hulls and those from wood; any difference being due to plant structure and pentosan content.^{7, 8, 9} If any appreciable difference were found, the economic status of these waste products might allow this type of utilization.

EXPERIMENTAL

The distillation was carried out in a cylindrical iron retort which was electrically heated from the outside by four 550-watt heating coils. The furnace was $2\frac{1}{2}$ inches in diameter by 18 inches long, inside dimensions, and insulated by a steel jacket containing $2\frac{1}{4}$ inches of magnesium-asbestos cement. Untreated hulls were introduced at one end of the core by removing an iron plate which was held against a half union by four $\frac{1}{2}$ -inch bolts. The whole was made air-tight by a copper-asbestos gasket between this iron plate and the half union connected to the core. The distillate was carried off by a $\frac{1}{2}$ -inch iron pipe connected through a pipe cap screwed to the other end of the core. With this apparatus it was possible to reach a red heat of iron within one hour, though more time was utilized in the process of distillation. The heating was continued until nothing more distilled over. Usually two or three hours' heating was required for each distillation.

The products distilled can be divided into three fractions by their physical properties. The gases were inflammable and contained carbon monoxide, carbon dioxide, hydrogen, and low molecular weight organic compounds. The liquid distillate was miscible with water, contained varied organic compounds to be discussed later, and large quantities of

⁶ Fred, Peterson, and Anderson, Ind. and Eng. Chem., **15**: 126 (1923).

⁷ Fraps, Am. Chem. J., **25**: 26 (1901).

⁸ *Wood Distillation*, by L. F. Hawley, Chem. Cat. Co. (1923).

⁹ *The Pyrolysis of Carbon Compounds*, by Charles Dewitt Hurd, Chem. Cat. Co. (1929).

water. The semi-solid portion which was immiscible with water and the water fraction, was colored a light yellow but darkened on standing. This color change occurred in the presence or absence of light and air. Also further tar was formed from the water fraction on standing. The

TABLE I
Compounds identified in pyrolygneous acid

| COMPOUND | DERIVATIVE | MELTING POINT | REMARKS |
|------------------------|---|---|--|
| | | °C. | |
| Acetone | 2-4 dinitrophenylhydrazone | 127 | Mixed M.P. 127°C. |
| Ethyl n-propyl ketone | 2-4 dinitrophenylhydrazone | 130 | Does not give test for aldehydes. Proven by mixed melting points not to be acetone or methyl n-propyl ketone |
| Methyl n-butyl ketone | 2-4 dinitrophenylhydrazone | 110.5 | Mixed M.P. 110.5°C. |
| Formic acid (solution) | No derivative. Quantity and dilution prohibited further treatment | Reduced KMnO_4 but does not give aldehyde test with Schiff's reagent, Fehling solution, or Benedict's solution. Precipitates HgCl_2 from HgCl_2 solution on heating. Solution regenerated from Ca salt and must be an acid | |
| Acetic acid | p-nitro benzyl acetate | 77.8 | Typical odor and proved to be acid. Mixed M.P. 77.8°C. |
| Ethyl alcohol | 3-5 dinitrobenzoate of ethyl alcohol | 92 | Mixed M.P. 92°C. |
| Butyric acid esters | No derivative. Quantity too small | NaOH saponification—acidification with H_2SO_4 gave unmistakable odor of acid | |

material used for further analysis was a composite of distillates, except gases, of many distillations. The carbon residue remaining in the furnace was quite finely divided and could easily be ground in a mortar to such a degree of fineness that colloidal solutions of the carbon would

TABLE II
Compounds indicated in pyroligneous acid

| | REMARKS |
|-------------------|---|
| Methyl Alcohol | Due to small volume of pure material, a pure derivative could not be obtained. Its odor and boiling point indicated the presence of methyl alcohol and by reactions the fraction was proved to be an alcohol |
| Allyl Alcohol | Dilute water solution. Reduced KMNO_4 and bromine in CCl_4 . Derivative was impure and reactions were not positive enough to be considered proper proof of the presence of allyl alcohol |
| sec-Butyl Alcohol | Dilute water-alcohol fraction, impure. No derivative. Water solution gave iodoform test. This is quite indicative after elimination of other compounds which give iodoform under these conditions, but is not considered definite enough to be conclusive |

TABLE III
Data on higher boiling fractions

| FRACTION | LITMUS | SOLUBILITY IN WATER | COLOR WITH FeCl_3 | SCHIFF'S | 2-4 DINITRO-PHENYL HYDRAZINE | NITROGEN | BOILING POINT |
|----------|-------------|---------------------|----------------------------|----------|------------------------------|----------|---------------|
| | | | | | | | °C. |
| 5 | Acid | Slight | Brown | Neg. | Neg. | Neg. | 106-118 |
| 6 | Acid | Slight | Reddish brown | Pos. | Neg. | Neg. | 146-150 |
| 7 | Acid | Slight | Reddish brown | Pos. | Pos. | Neg. | 150-155 |
| 8 | Acid | Slight | Reddish brown | Pos. | Neg. | Neg. | 157-160 |
| 9 | Acid | Neg. | Dull brown | Pos. | Neg. | Pos. | 160-190 |
| 10 | Acid (weak) | Neg. | Brown | Pos. | Neg. | Pos. | 198-205 |
| 11 | Acid (weak) | Neg. | Brown | Pos. | Neg. | Pos. | 205-210 |
| 12 | Acid (weak) | Neg. | Brown | Neg. | Neg. | Pos. | 212-200 |
| 13 | Acid (weak) | Neg. | Brownish black | Neg. | Neg. | Pos. | 220-225 |

result when it was treated with water. No significant work was done on the carbon. This will be studied later.

The pyroligneous acid or water fraction was treated with lime to

remove the acids and then subjected to fractional distillation. The methyl ketones were then separated by sodium bisulfite. By thus separating this fraction of the distillate, some identifications have been made, as shown in Table I. Due to the large excess of water, the number of products, and the limited quantity of material at hand, many compounds that were evidently present were not definitely proved as constituents of the distillate. However, the results from the physical properties and chemical reactions have rather definitely proved that the products were quite similar to the products of wood distillation, if not somewhat more complex. The evidence concerning the presence of some other compounds not definitely proved to be present is discussed in Table II.

Termination of the research left the oil fractions without identifications. However, the oils were fractionated and many reactions determined. This oil contains fractions which boil between 100°C and 225°C. Together with these boiling points are given some of the properties of the individual fractions of this oil, Table III.

The tar product was a plastic solid at ordinary temperatures and contained small amounts of carbon. Since no indications of aldehydes were found throughout the work, it is logical to conclude that the tar is formed from these compounds and phenols, along with other polymerized and high boiling products.

CONCLUSIONS

This mode of destructive distillation resulted in products little different from those derived by destructive distillation of wood. The bulk of this type waste product will also be an inhibiting factor in this method of utilization. The work as a whole has shown that the products are identical in physical properties with those from wood distillation, that their chemical content is approximately the same, and that such a process would inherit all the difficulties known in this industry today. These difficulties center themselves in the complexity of the materials obtained. Under our present economic status of synthetic chemistry, petroleum, and coal, this type of process has little chance of existence. In case of a shortage of coal or petroleum, this material can be utilized as a source to obtain similar products as substitutes. The research program has indicated other possible avenues for utilization of these products but opportunity to follow them up has not arisen.

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ACHLYA FLAGELLATA AS A FISH PARASITE

By WESLEY N. TIFFNEY and FRED T. WOLF

In the spring of 1935 the senior author observed in a small pond near Lexington, Massachusetts, a considerable number of newts infected with a fungous disease. The host was identified as *Triturus viridescens*. Specimens were brought into the laboratory, and two species of fungi, *Saprolegnia parasitica* Coker and *Achlya flagellata* Coker,¹ were isolated from a number of them. The presence of both species of fungi on the infected animals was furthermore verified by direct examination. Inasmuch as *Achlya flagellata* occurred in connection with *Saprolegnia parasitica*, which is known to be a severe parasite, while *Achlya flagellata* has apparently never been reported in the literature as a parasite, little significance was attached at that time to the isolation of the latter species from infected animals.

The second isolation of *Achlya flagellata* from diseased animals occurred in the autumn of 1935. Specimens of *Lebistes reticulatus* had been secured and kept in a 25-gallon aquarium tank in water from the Cambridge, Massachusetts, municipal supply. These fish were intended for use in a study which was being made of some physiological aspects of parasitism by *Saprolegnia parasitica*. After about eight weeks in the aquarium tank, twelve of the seventy fish were observed to be infected by a fungus which was identified as *Achlya flagellata*. No other species of water mold was present, although large masses of bacteria appeared about the infected areas on the fish. From experience gained in studying the etiology of the *Saprolegnia parasitica* fish disease, the senior author believes that bacteria, which usually are not fatal to the fish, should be regarded as secondary invaders. All the fish infected by the fungus died.

Achlya flagellata was isolated from the diseased specimens of *Lebistes reticulatus* and grown on hemp seed in sterile distilled water; the bacteria-free culture was used to inoculate *Fundulus heteroclitus* in an attempt to verify the ability of the fungus to act as a parasite and to determine whether injury to the host had any effect on the incidence of

¹ Coker, W. C. 1923. The Saprolegniaceae with notes on other water molds. 116-118, pl. 37. Univ. of N. C. Press, Chapel Hill.

infection. The 60 fish used in this experiment were divided into three groups as follows: Group I consisted of 25 fish which were injured slightly by the removal of a few scales, Group II consisted of 25 uninjured fish, and Group III included 10 fish, of which 5 were injured and 5 uninjured. Each group was placed in a separate tank. All tanks received water from the same source, and were maintained under identical conditions of temperature, light, and aeration. Groups I and II were exposed to zoöspores of *Achlya flagellata*, while Group III, which was not exposed to the fungus, served as a control. Of the 25 injured fish in Group I, 9 were attacked by the fungus and died. In no case did infection result in the uninjured fish (Group II) or the controls (Group III). This experiment apparently places *Achlya flagellata* in the category of wound parasites.

During the early summer of 1936, *Achlya flagellata* occasioned severe losses in the Bayfield, Wisconsin, hatchery of the state Department of Conservation. According to Mr. B. O. Webster, Superintendent of Fisheries, who provided much of the information concerning the epidemic, there had been no history of outbreaks of fungus disease at the Bayfield hatchery in past years, nor did the disease occur to the extent of economic importance in other hatcheries in Wisconsin during 1936.

At the Bayfield hatchery, about 3,000,000 fingerlings of brook trout, *Salvelinus fontinalis*; rainbow trout, *Salmo irideus*; and brown trout, *Salmo fario*, were raised in metal tanks measuring three feet by fourteen feet by eight inches. The water used comes from two sources; the first is an artesian well, and the second is a creek called Birch Run. No infection occurred in the tanks supplied with water from the former source. For this reason, after the epidemic had become established, water from the artesian well was used in all of the tanks, and common salt was used liberally in an attempt to control the disease, but these measures were applied too late to be of much value.

The first cases of infection appeared late in May. On May 29, Dr. E. M. Gilbert collected samples of water from the tanks, masses of fish food overgrown with the fungus, and infected fish. Hemp seed cultures made from these collections by the junior author showed that *Achlya flagellata* was the cause of the disease; no other fungi were involved. Dr. J. N. Couch kindly identified the fungus from subcultures from the infected fish.

It was found that great differences were displayed by the three species of trout in regard to resistance to infection by the fungus. Fingerlings of brown trout, *Salmo fario*, and rainbow trout, *Salmo irideus*, appeared

to be almost immune to the disease. With brook trout, *Salvelinus fontinalis*, however, about 50 per cent of the fingerlings, representing about 500,000 fish, were killed. The percentage of infection was slightly larger than this figure would indicate, as some of the infected fish recovered. Although the disease was most severe in its attack upon the fingerlings, larger fish were also infected with the fungus and killed. The epidemic ran its course, and was well under control by the middle of July.

It appears, therefore, that *Achlya flagellata* may at times become a serious parasite of fish under Wisconsin conditions. In Massachusetts, on the other hand, the senior author has encountered it only twice within a period of three and a half years in a series of 128 isolations of fungi from diseased fish found under natural conditions. The details of these two collections have been given above. It would appear that *Saprolegnia parasitica*, found 122 times, is the predominant parasitic form in that region, and that *Achlya flagellata*, although interestingly enough capable of parasitism, is of little importance there.

The work of the senior author was carried out in the Laboratories of Cryptogamic Botany of Harvard University, under the direction of Dr. W. H. Weston, Jr., while that of the junior writer was done in the Department of Botany, University of Wisconsin, and supported by a grant from the Alumni Research Foundation. To the various persons who have been of assistance in this investigation, the writers wish to express their appreciation.

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THE FORMATION AND OPERATION OF THE TRAPS IN THE NEMATODE-CATCHING FUNGUS, *DACTYLELLA* *BEMBICODES* DRECHSLER

By JOHN N. COUCH

PLATE 27

The recent papers by Drechsler (1933, 1937) have greatly added to our knowledge of the nematode-catching fungi and to an understanding of the means by which the nematodes are trapped. No experimental observations, however, have been reported which describe the conditions under which the traps are formed or how they close or the stimuli which may cause their closure. Is the formation of the traps conditioned by the presence of nematodes? Do the traps close instantaneously or slowly? Is closure brought about by a mechanical or chemical stimulus? The chief objects of this investigation have been to discover the conditions which determine the formation of the nematode traps in *Dactylella bembicodes* and how they operate in catching the nematodes.

The present fungus was found growing over a small piece of decorticated, semi-decayed, pine wood on which there had been some *Dacryomyces* material. It was first recognized under the binocular microscope by the tiny, upright, solitary conidiophores. Upon mounting some of the mycelium, numbers of nematodes, some living, others dead, were found attached to the fungus by small traps.

STRUCTURE OF FUNGUS

The fungus produces a rather scanty growth on the wet wood, but a heavy mycelial growth is formed on certain nutrient agars. The threads are rather straight, branched, frequently septate, $3.8-4.6\mu$ thick, with a considerable number of the H type of anastomoses. Conidiophores borne abundantly in most cultures, thick at base, about $2.8-3.5\mu$ thick near tip and $400-1500\mu$ tall, with one apical spore. The spore when mature is cut off from the stalk and may either fall off or be carried away by air currents. Spores ovoid, bluntly pointed at both ends, thickest in the distal part of the middle region, four-celled with a small distal, two small proximal, and a large central cell, in which

is a large, glistening globule of material. When growing on moist wood or in aqueous solutions with nematodes, the fungus forms numerous small rings or loops; rings are also produced on certain kinds of nutrient agars, and when the food material is reduced. Each loop consists of three cells and is attached to the hypha by a short, two-celled stalk. In the formation of the loop the last loop-cell anastomoses with the top stalk-cell and also with the first loop-cell. Loops $24-31\mu$ outside diameter, $16-21\mu$ inside diameter. The spores germinate readily, producing a germ tube of even diameter about $3.6-4\mu$ thick. Most commonly a germ tube arises from each end of the spore, growing out straight in a line parallel to the long axis of the spore. Sometimes a germ tube may arise from the small cell next to the end cell, the tube growing out at about right angles to the spore. When the spores germinate in a liquid medium containing nematodes, three or four rings are formed on the germ tube before it has attained a length of 300μ .

Some preliminary observations were made to determine how the rings functioned in catching the nematodes. Some fungal threads bearing rings were isolated on a slide in water with several nematodes and it was possible under the binocular in a few instances to maneuver the nematodes into the loops. Because of the low magnification, however, it was impossible under the binocular to discover how the trap worked and, in every instance observed, before the mount could be transferred to the compound microscope the traps had completely closed and the nematodes were held fast. Moreover it was impossible to observe the traps under the compound microscope with the fungus growing over the surface of the wood.

To overcome these difficulties the fungus and nematodes were cultured in nutrient agar.

ISOLATION OF NEMATODES IN CULTURE

In culturing the nematodes the technique described by Chandler (1924) was used at first. Chandler used "ordinary nutrient agar" with a drop or two of dirty water to supply bacterial growth. Dirty water, however, almost invariably contains the spores of various fungi the growth of which renders such cultures worthless. Therefore maltose peptone agar plates inoculated with one or more species of bacteria from pure cultures were tried on which to culture the nematodes. The nematodes grew well for several days and then died out, perhaps due to the accumulation of waste products in the agar. After some further experimentation it was found that by adding some living cells of the green

alga, *Chlorella*,¹ to the bacterial inoculation the nematodes multiplied abundantly and remained healthy and active for weeks or until the agar culture dried up.

ISOLATION OF FUNGUS IN PURE CULTURE

The fungus was easily obtained in pure culture since the spores are large and germinate readily on nutrient agar. A place on the original culture on wood where the conidiophores stood out distinctly and were free from trash was located under the binocular microscope. With a needle to which was attached a small block of agar, a single spore could be picked up by touching the spore to the surface of the agar. The little block of agar to which the spore was attached was transferred to a sterile maltose-peptone agar plate or tube.

EXPERIMENTS TO INDUCE THE FORMATION OF RINGS

The fungus grew slowly on maltose-peptone agar No. 5, the largest colonies attaining a diameter of 15 mm. after ten days. However, on the maltose peptone agar no rings were formed. Since numerous rings were formed when the fungus was growing over the surface of wood, it was suspected that the wood might contain something which induced their formation. The fungus was then grown in a sterile hay decoction but no rings were formed. However, a drop of the brownish water from the original dish containing nematodes and fungus placed on the edge of the mycelium on maltose peptone agar caused the formation of numerous rings in that region after twenty-four hours. The water in the original culture was tested and found to be acid. Suspecting that the acidity of the medium might influence the formation of the rings, cultures of the fungus were made on maltose peptone agar to which were added varying amounts of H_3PO_4 with results as indicated in the accompanying table. The cultures were observed occasionally during a period of two weeks.

| | |
|--|------------|
| 1. 0.1 cc. of 0.1N H_3PO_4 in 20 cc. agar No. 5..... | No rings |
| 2. 0.2 cc. of 0.1N H_3PO_4 in 20 cc. agar No. 5..... | No rings |
| 3. 0.5 cc. of 0.1N H_3PO_4 in 20 cc. agar No. 5..... | No rings |
| 4. 1 cc. of 0.1N H_3PO_4 in 20 cc. agar No. 5..... | Many rings |
| 5. 2 cc. of 0.1N H_3PO_4 in 20 cc. agar No. 5..... | Many rings |
| 6. 3 cc. of 0.1N H_3PO_4 in 20 cc. agar No. 5..... | Many rings |

¹ Cultures of *Chlorella* have been kept going in the laboratory for several years on Bristol-Roach agar (see Ann. Bot. 40: 149. 1926).

This experiment indicated that by acidifying the culture media to a certain degree the fungus could be induced to form many rings. The rings are formed only on the hyphae which grow on the surface of the media and stand upright with their faces at right angles to the hyphae from which they arise. In later experiments it was found that growth was about twice as luxuriant and the production of rings more abundant on Blakeslee's No. 230 agar than on acidulated maltose peptone No. 5. On potato dextrose agar (Chamberlain, *Methods in Plant Histology*) growth was about twice as luxuriant as on No. 230 but no rings were formed.

Also it has been noticed that the production of rings is greatly increased as the available food supply in the culture media is reduced. The production of rings occurs on agar No. 230 but when the mycelium reaches the edge of the dish or when a groove is cut in the surface of the agar at the edge of the growing region, the number of rings is greatly increased. Furthermore when several inoculations are made on the same culture dish and the mycelium grows together the same increase in ring production occurs. It thus appears that the traps may be formed as a response to a reduced food supply.

OBSERVATIONS TO DETERMINE HOW THE TRAPS WORK

With the fungus in pure culture producing abundant rings and with plenty of nematodes also at hand, it was possible to carry out some experiments to see how the rings work.

The following observation was made to determine if the rings or traps close instantaneously or if they close by a slow swelling (i.e. taking a few seconds to close) of the three cells. This observation had been impossible before with the fungus growing on wood where observations with a compound microscope were worthless. Agar teeming with nematodes was scraped off and placed on the center of a fungus culture on which there were numerous rings. The following morning, about twelve hours later, the colony was observed. Many nematodes had wriggled from the central mass of agar and were crawling about over the surface of the agar over and among the fungal hyphae. Many nematodes had already been caught in the traps. Some were still actively wriggling in a futile effort to free themselves, others had already been killed and their bodies invaded by the fungus. Attention was concentrated on one active, free nematode gliding sinuously about among several rings. For a while it would move rapidly forward or again it would stop and thrust its head forward first in one direction, then in another. After a time the nematode thrust its head into one of

the rings, upon which the three component cells of the ring immediately swelled gripping the worm so firmly that it could neither move forwards nor backwards. It did squirm violently, however, in a futile effort to free itself. Once the nematode is caught in the trap its death is certain, for in no instance has one been observed which was able to free itself. Using the above technique a number of nematodes have been observed to enter the rings and in all cases when the worms are caught the action of the trap is instantaneous.

In some instances all the nematodes may be caught in the traps. Usually, however, some of the nematodes miss the traps and crawl out on the clean agar carrying bacteria with them. These will lay eggs in the agar so that new nemas will be coming on. Meanwhile the bacteria and *Chlorella* multiply, the former to furnish food for the nemas. The fungus continues to grow and form new rings which in turn trap some of the new nemas. Such cultures consisting of the fungus forming rings, the nemas, bacteria, and *Chlorella* may keep going for several days. As a rule in such cultures the bacteria get the upper hand, the fungus overrun by bacteria stops growing and the rings lose their ability to function. In such old cultures, upon several occasions, I have observed a nematode crawl through a ring and even stop and loiter in it for several minutes with impunity. The rings appeared perfectly normal but for some reason had lost their ability to close.

If the nematodes are slow to enter the rings, it is possible to hasten the process by pushing them with needles, using a high powered binocular dissecting microscope. The nema shown in figure 16 was pushed into position and entered the loop (a), the cells of which swelled immediately holding the nema fast. The thread (t) to which this loop was attached was then cut leaving a long end in front. Then by holding the thread (t) on a needle the nema was pulled over the agar and its head slipped into another loop (b). In spite of the fact that the nema was still alive and vigorous this ring failed to swell even after an hour. Meanwhile the nema's tail end was threaded into a third loop (c) which, however, also failed to close. It is very likely that the loops (b) and (c) were injured in manipulation, for numbers of cases have been seen where a nematode was caught by two loops arising from the same or from different hyphae.

EXPERIMENTS TO DETERMINE WHAT CAUSES THE LOOPS TO SWELL

Having observed that the nematodes were caught by the instantaneous swelling of the three loop cells thus closing the loop, it seemed of great interest to discover what caused the cells to swell.

The vigorous action of the nematodes suggested the idea that the closure of the trap might be due to the mechanical irritation brought about by the vigorous movements of the nematode's body. To test this theory a fine glass rod was inserted into a loop under a high power binocular dissecting microscope and the rod moved back and forth in the loop, simulating as much as possible the movements of a nematode. Later the same experiment was performed several times with a Fitz microdissection apparatus but in no case could any more than a slight swelling be obtained (fig. 10). It was apparent from these experiments that mechanical irritation plays a very small, if any, part in the closure of the loops.

The above experiments were made before I had seen one of the traps close on a nematode under the compound microscope. Here one can see that the violent movement of the nematode takes place after it is caught, not while it is being caught, for that occurs instantaneously. It seemed likely, therefore, that the stimulus causing the closure of the traps might be of a chemical nature. Accordingly weak solutions of ammonia, potassium hydrate, phosphoric acid, acetic acid, hydrochloric acid, sulphuric acid, and lactic acid were applied to the loops. Treated with a 1% solution of lactic acid the loops were caused to swell slightly but in none of the experiments could complete closure be obtained. These experiments were inconclusive, failing to show positively that the closure of the traps was due to chemical stimulus.

That the closure of the traps is due to a chemical (as opposed to mechanical) stimulus was discovered accidentally and in a very round-about way. It was noticed on many occasions that when blocks of agar with hyphae were cut from the pure culture the loops near the cut edges were always swollen. It was suspected at first that cutting the hyphae to which the loops were attached might permit a sudden intake of water and the consequent swelling of the loops. However, upon cutting some hyphae isolated on a slide in a block of agar the loops all remained unswollen regardless of how badly the hyphae to which they were attached were mutilated. It then occurred to me that I was using a cold scalpel whereas a hot one was invariably used to cut the blocks from the stock cultures. Could heat cause the cells to swell and the traps to close? To answer this a block of agar with threads on which were many loops was cut from the stock culture, placed on a slide and a hot scalpel held about two millimeters above the surface for about two seconds. The loops were then examined under the microscope and all of them excepting a few were completely closed (as in fig. 9). Indeed it was possible

to locate several loops under the low power of the microscope and watch them close by having some one wave a hot needle over the loops. Such experiments have been repeated many times with the same results. It is interesting to note that only the three cells of the loop swell; the cells of the hyphae and the spores are not visibly affected by the heat.

To determine how much heat was necessary to cause the swelling, warm, distilled water was dropped on the loops. It was found that water at any temperature from 33°C.-75°C. would cause the loops to close. Below about 30°C. there was no swelling and above about 80°C. the cells failed to swell being killed by the heat.

An examination of the cells which have been caused to swell by dry heat or warm water shows that the three swollen cells contain a refractive substance which is doubtless gelatinous or colloidal in nature. The closure of the trap is due to the sudden swelling of this substance. The heat doubtless "activates" the colloids causing them to swell. This swelling may be due to a rearrangement of the molecules of water and colloidal material already in the cell or it might be that additional water is imbibed from the stalk and thread cells. The rapidity of the reaction would seem to favor the first view.

The closure of the traps by heat does not throw much light on how the nematodes caused their closure. It is assumed that the temperature of the nematode's body is that of other cold blooded animals and therefore it is certainly not the "heat" of the nematodes' body that throws the trap. It can hardly be doubted, however, that the nematode gives off some substance which diffuses into the loop cells causing the swelling and the consequent closing of the trap.

After a nematode is caught in a trap its body is rapidly invaded and filled by the fungal hyphae with the consequent consumption of the nematode as food by the fungus. New hyphae sprout out from the parasitized nema's body and extend out over the substratum giving rise to more traps and one or more conidiophores.

Since the fungus grows readily in artificial culture, producing abundant spores, it is possible that the spores might be mixed with soil and broadcast over small areas of nematode infested soil with the consequent control of the nematodes (see Linford, 1937).

SUMMARY

A fungus that catches nematodes is described. The fungus has been isolated in pure culture and grown on a variety of media. It has been found that the nematode traps are produced when the fungus is grown

on acid media. Also a reduction in the food supply induces the formation of rings, as when the fungus reaches the edge of the culture dish or when a groove is cut in the agar at the edge of the growing region. On certain culture media, e.g. potato dextrose agar, the rings are not formed at all.

The nematodes have also been isolated and grown in culture on agar. To furnish food for the nematodes the agar plates were inoculated from pure cultures of bacteria. The addition of the green alga, *Chlorella*, to the cultures of nematodes keeps the cultures in a healthy condition for a longer time.

The closure of the rings has been observed under the compound microscope. When a nematode thrusts its head or tail into one of the rings it closes practically instantaneously by the simultaneous swelling of the three cells of the ring.

Partial closure of the rings may be induced by mechanical irritation of the ring as when a fine glass rod is inserted and moved back and forth in the loop. Also slight closure may be induced in a few rings by a 1% lactic acid solution. Numerous other dilutions of acids and alkalies failed to cause any swelling. Complete and instantaneous closure may be induced by the application of heat, either dry heat or warm water or steam. Using warm distilled water it was found that a drop or two of water of any temperature from 33°C to 75°C applied to rings caused instantaneous and complete closure. In such experiments only the cells of the rings swell, the threads being unaffected. Water above 80°C failed to cause swelling, the high temperature killing the cells and perhaps inactivating the colloids. A microscopic examination of the swollen loop cells showed that they contained a refractive gelatinous looking substance, the substance doubtless which had been caused to swell.

The experiments with heat demonstrate that the swelling may be initiated by a stimulus such as heat but at present it is impossible to see the relation between the closure of the traps by heat and by the "cold blooded" nematodes. It is assumed that the living nematodes give off a chemical substance which induces the swelling of the colloids just as heat induces their swelling.

After the nematode has been caught, the fungal hyphae invade and destroy the animal's body, using it as food.

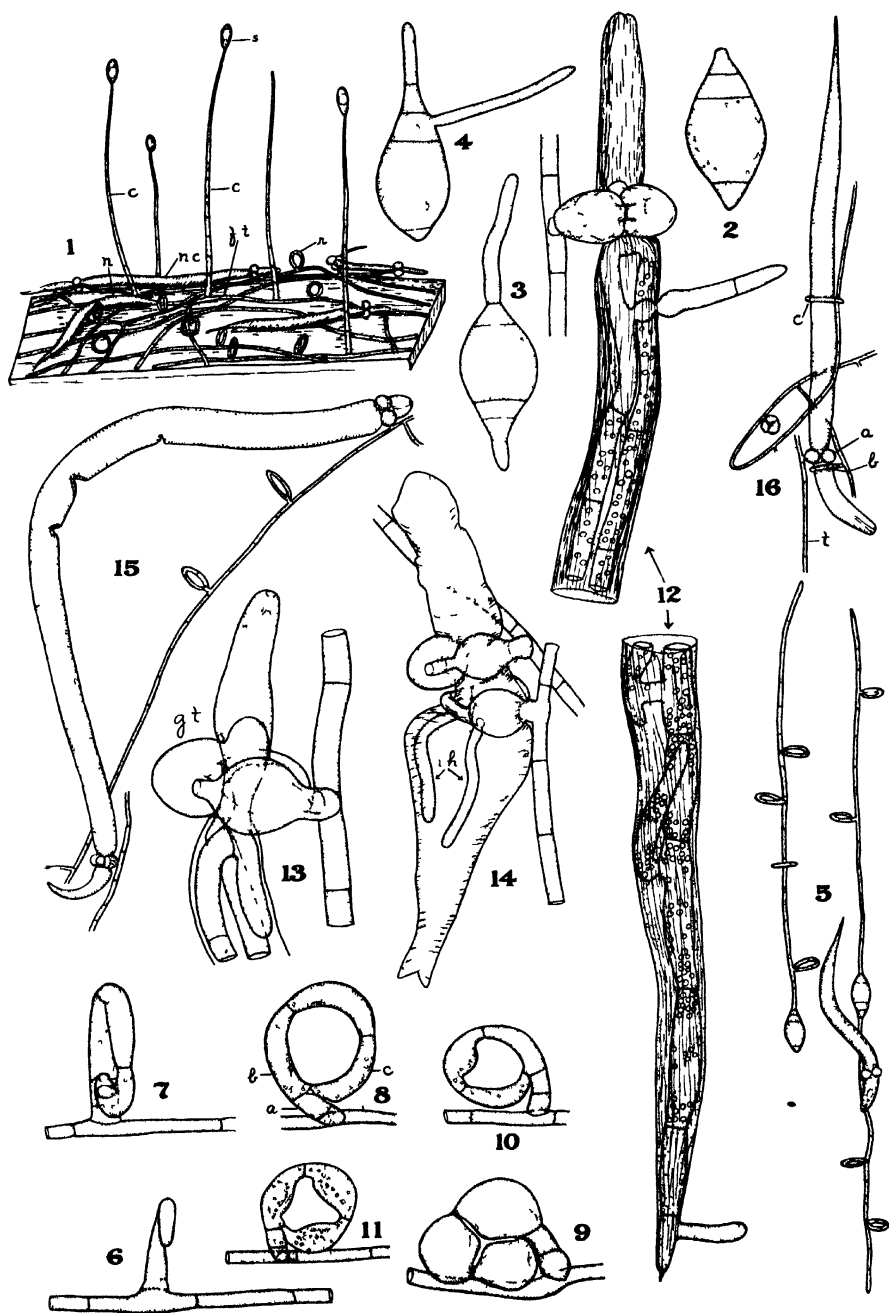
The suggestion of biological control of soil nematodes by the fungus is given.

LITERATURE CITED

- CHANDLER, A. C. Free-living Nematodes. *Science* **60**: 203. 1924.
DRECHSLER, CHARLES. Some More Fungi That Prey on Nematodes. *Journ. Wash. Acad. Sci.* **23**: 355-357. 1933.
———, Some Hyphomycetes that prey on free-living terricolous Nematodes. *Mycologia* **29**: 447-552. Fig. 1-18. 1937.
LINFORD, M. B. Stimulated Activity of Natural enemies of Nematodes. *Science* **85**: 123-124. Jan. 29, 1937.

EXPLANATION OF PLATE 27

- Fig. 1. Diagrammatic habit sketch of fungus growing over a damp piece of decayed pine wood, showing fungal threads (f.t.) on which are formed the rings (r) which catch the nematodes (nc), also conidiophores (c) on which the spores (s) are formed. $\times 50$.
Figs. 2-5. Germinating spores. In fig. 5 the hyphae floating on the surface of water have formed several nematode traps one of which has caught a nematode. $\times 110$.
Figs. 6-11. Formation and structure of fungal rings.
Fig. 6. Lateral branch growing out, the tip curving back, as it grows to fuse eventually with the upper stalk cell (fig. 8, a) and the first loop cell (fig. 8, b). Figs. 7 & 8. Edge and side views of mature loops. Fig. 9. A loop the three cells of which have expanded throwing the trap without catching a nematode. Fig. 10. A loop into which was inserted a glass needle, by the vigorous movements of which one of the cells was caused to swell. Fig. 11. One of several loops treated with 1% lactic acid. All $\times 525$.
Fig. 12. Above, the anterior, below, the posterior part of a trapped nematode, the body of which has been invaded by fungal hyphae. $\times 580$.
Fig. 13. Germ tube (g.t.) from one of loop cells penetrating body wall of nematode to form the invading hyphae. $\times 930$.
Fig. 14. Large rotifer trapped and invaded by fungus. i.h. infecting hyphae. $\times 525$.
Fig. 15. Large nematode trapped by both head and tail ends. This worm's body was completely filled with fungal hyphae. $\times 110$.
Fig. 16. Nema shown was maneuvered into loop (a), i.e. pushed into position and entered loop itself, the cells of which immediately swelled holding nema fast. See text for further explanation. $\times 110$.



ADDITIONS, CORRECTIONS, AND DELETIONS FOR THE FLORA OF THE TRYON REGION

By DONALD CULROSS PEATTIE

In the spring of 1919 I began the study of the flora of the Tryon region of Polk County, North Carolina, and some adjacent regions, chiefly mountainous, in South Carolina. After collecting many hundreds of specimens I began to search the leading herbaria for previous collections from this region and entered into correspondence with other collectors. Some were kind enough to make at my request special collections of interesting genera when they went to the Tryon country. The results were finally embodied in a series of papers entitled "Flora of the Tryon Region of North and South Carolina: An annotated list of the plants growing spontaneously in Polk County, North Carolina, and adjacent parts of South Carolina, in Greenville and Spartanburg counties." It appeared in six parts, running in the Journal of the Elisha Mitchell Scientific Society (Chapel Hill, N. C.) from September, 1928, to June, 1931 (vols. 44-46).

Between 1926 and 1936 I did not revisit this region, but in that time a few species were added to the flora by other collectors, notably Miss N. B. Kimber who found additional species of *Asarum*, *Phlox*, and *Asplenium* and Mr. B. E. Smith who added two dodders to the known flora. Notable, too, was the rediscovery on the north side of Tryon Mt.* of the Rocky Mountain *Woodsia* by Dr. Edgar T. Wherry. *Woodsia scopulina* had first been found in this state in 1897 at that locality by E. C. Townsend, but had never since been located.

The spring flora of the Appalachians is well known, owing to its beauty and the number of visitors who come at that delightful time of year, but little attention is usually paid to the autumn flora, which bristles with the technical difficulties of asters and goldenrods and other composites. It was with great interest, therefore, that in September of 1936 I took a house in the country outside the town of Tryon and prepared to see what could be added to the known list of 1076 vascular plants. Diligent collecting was all that was needed to add many autumn-

* Called White Oak Mountain in Wherry's note (Amer. Fern Journal 24: 101. 1934). Ed.

nal Compositae. But more surprising were the following additions which are also, I believe, new to western North Carolina: the water-weed, *Anacharis densa*; the African grass, *Eulalia viminea*; *Trillium declinatum*; a Louisiana violet, *Viola Lovelliana*; and, most astonishingly, the salt marsh shrubby composite, *Baccharis halimifolia*.

I wish to acknowledge gratefully the assistance, in the naming of many of my collections, of Mrs. Agnes Chase, who identified the grasses, Dr. Edward Palmer who named the woody plants, Dr. N. C. Fasset, who examined all aquatics, Dr. S. F. Blake who identified the Compositae, and Dr. John K. Small who named several miscellaneous species that were puzzling. My specimens have all been sent to the herbarium at the University of North Carolina, except for the woody plants, which remain at the Arnold Arboretum. My collections extended from Sept. 28, 1936, to May 16, 1937. Most of the new species were, naturally, autumnal, yet the spring flora yielded thirteen species, including three species of the difficult brambles.

It is my pleasure, also, to acknowledge that certain species new to the flora were drawn to my attention by resident amateurs. Mrs. George Holmes showed me where the checkerberry, *Gaultheria procumbens*, grows on Godshaw Hill, apparently the only locality in the county. Miss Helen Stearns, in her garden, showed me the handsome clubmoss, *Lycopodium complanatum*, which she had brought down from Melrose Mt. And Mr. William Weigel, an enthusiastic wildflower gardener, has transplanted to his plot the monkshood, *Aconitum uncinatum*, and the old favorite, lily-of-the-valley, which grows wild on Rocky Spur, in Greenville Co., S. C. Native to Europe and the mountains of Asia, this is also an inhabitant, as an indigene, of the southern Appalachians, but it was new to the recorded flora of the Tryon Region.

A special study was made of *Asarum*, a genus which reaches its highest development in the southeastern states, in order to test the validity of the many species that have been proposed and often all too loosely described. With the exception of *Asarum Memmingeri* which was not found this time, all the species hitherto reported from this region were located again, collected in quantity and very carefully studied. It is the conclusion of this writer that they are better founded than one would judge from the descriptions of them in books.

It was a pleasure also to be able to add two species and one new variety to the recorded trilliums of the region. Several trees and shrubs were also newly discovered, giving this county one of the longest and most interesting lists of woody plants in the state.

The present writer is now leaving for a few years in the West. He hopes that others will investigate the peculiarly rich flora of this region where the piedmont meets the front wall of the Blue Ridge and so many diverse elements are met together. Further collection should certainly greatly increase the number of grasses and sedges. The mountains in the northwestern corner of the county have never been botanically investigated by anyone, so far as I know. The extreme eastern edge of the county has only been entered by a botanist on a single occasion. The size of the "unknown" area cannot, of course, be taken as a measure of the possible "unknown" plants, since the well-known regions sufficiently resemble the little-known in their vegetation to make it likely that the present number of some 1155 species of flowering plants and ferns could not be increased by much more than another hundred species. Most of these will undoubtedly be found among the highly technical groups.

ADDITIONS

FERNS

Asplenium pinnatifidum Nutt. (Spleenwort)

This interesting little plant, which so much resembles the walking fern, has been found on cliffs and rocks in Greenville Co., S. C. Specimens: Hogback Mt., Feb. 12, 1930, and Clarke's Mt., March 2, 1930 (Miss Kimber).

Selaginella apus (L.) Spring.

Much resembling a moss, this little fern ally appears to be quite common on wet rocks near waterfalls and brooks. I found it in quantity in Vaughan's Glen. Specimen: Dec. 1, 1936. No. TR19.

Lycopodium complanatum L. (Trailing Christmas Green)

This handsome little plant grows on Melrose Mt. Miss Helen Stearns has transplanted a quantity of it to her garden.

EEL-GRASS FAMILY

Anacharis densa (Planch.) Vitt. (Waterweed)

A tropical plant native of South America which has previously been recorded only from Florida. Specimen: Lake Lanier, Oct. 21, 1936. *Fide*, N. C. Fassett. No. TR122. Lake Lanier is in Greenville Co., S. C.

GRASS FAMILY

Eulalia viminea (Trin.) Ktze.

Adventive from Africa, this grass seems to have made its entry through Norfolk, Virginia, whence it has spread southward. It is of such recent advent that it is not noted in Small's new Manual. Specimen: fields and roadsides, Oct. 7, 1936. No. TR34. *Fide*, Agnes Chase.

Eragrostis hirsuta (Michx.) Nees.

A roadside grass. Specimen: Sept. 28, 1936. No. TR20. *Fide*, Agnes Chase.

Eragrostis spectabilis (Pursh) Steud.

A beautiful grass. Specimen: In deep mountain woods, Oct. 4, 1936. No. TR98. *Fide*, Agnes Chase.

Sorghastrum Elliottii (Mohr.) Nash. (Indian Grass).

Specimen: Sunny mountain slopes, Oct. 10, 1936. No. TR61. *Fide*, Agnes Chase.

SEDGE FAMILY

Carex gynandra Schweinitz

Specimen: By a stream in a forest at the old Poinsett bridge, foot of Calahan Mt., May 13, 1937. No. TR186. This locality is in Greenville Co., S. C.

Carex hystericina Muhl.

Specimen: Beside the Saluda River at Merrittsville, May 13, 1937. No. TR187. This locality is in Greenville Co., S. C.

LILY FAMILY

Convallaria majalis L. (Lily-of-the-valley)

This beautiful little flower was shown me in bloom in Mr. William Weigel's garden. He brought his plants from the west slopes of Melrose Mt.

Trillium Underwoodii Small

This plant, intermediate in many ways between *T. simile* and *T. Hugerii*, has undoubtedly been here all along, but was passed over. On March 24, 1937, I took a satisfactory specimen of it, however, adding a new *Trillium* to the already impressive local array of this genus. Specimen: No. TR190.

Trillium grandiflorum (Michx.) Salisb.

In the *Flora of the Tryon Region* I reported only the variety *parvum* of this species. Miss May E. Day of Oberlin, Ohio, called my attention to the fact that the true species also flourishes here, and in 1937 I found plenty of it, and none of var. *parvum*. This makes one wonder if the peculiar little variety is not merely a depauperate phase thrown up by the plant in years somehow unfavorable.

Trillium declinatum (Gray) Gleason

I had never previously seen specimens of this species from North Carolina, and did not report it in my monograph of the genus in North and South Carolina. The range as described in Small's new Manual seems not to include the Carolinas. All that I found were in a single locality, at Bear Creek, Buck Mt. The flowers are deliciously fragrant, but not like those of any other *Trillium*. With its white ovary, rose-purple stigmas, and lilac anthers it is a striking flower. Specimen: April 26, 1937. No. TR168.

Trillium Vaseyi Harbison.

The only specimens were seen May 15, 1937, at Pearson's Falls. As this is now a wildflower preserve it was not possible to take away any plants. But of course there can be no doubt about the striking flower which looks much like *Trillium simile* but with curled sepals, maroon-rose petals, and declined or horizontal flower. The odor of roses was marked.

ORCHID FAMILY

Spiranthes cernua (L.) Richard. (Ladies' Tresses)

By shady mountain springs. Specimen: Oct. 14, 1936. No. TR140.

BIRCH FAMILY

Corylus cornuta Marsh. (Beaked Hazelnut)

Corylus rostrata Ait.

On high ridges, Rocky Spur. Specimen: Nov. 2, 1936. No. TR140.
Fide, Edw. Palmer. Rocky Spur is in Greenville Co., S. C.

KNOTWEED FAMILY

Rumex obtusifolius L. (Dock)

Common weed of European origin. Specimen: Oct. 7, 1936, by a spring, Whiteoak Mt. No. TR88.

Polygonum hydropiperoides Michx. (Mild Water Pepper)

In a brook, Valhalla. Specimen: Sept. 28, 1936. No. TR18.

PINK FAMILY

Dianthus Armeria L. (Deptford Pink)

I have no specimen of this little European waif, owing to the fact that I noticed it only while waiting to board the train to leave the Tryon region, May 28, 1937. It was present in some abundance in grass, and is of course an unmistakable species. It will undoubtedly spread, as it is naturalized elsewhere in the neighboring counties and should bloom all summer.

WATER-LILY FAMILY

Nymphaea odorata Ait. (Fragrant Water-lily)

Common in the shallow west bays of Lake Lanier. Specimen: Oct. 21 (still in flower), 1936. TR115. *Fide*, N. C. Fassett.

BUTTERCUP FAMILY

Aconitum uncinatum L. (Clambering Monkshood)

Mr. William Wiegel, who gave me my specimen (No. TR188) from his garden, collected this on dry hills near Lynn. We searched for it, but could not find it in May. It blooms, of course, in autumn.

STRAWBERRY SHRUB FAMILY

Calycanthus fertilis Walt. (Strawberry Shrub)

In the *Flora of the Tryon Region* I reduced this species to a synonym of *C. floridus*, but I am convinced on further investigation that this is a valid species and I was in error. It is common around Tryon.

MUSTARD FAMILY

Draba verna L. (Whitlow Grass)

This very precocious little lawn weed, adventive from Europe, is now established in many places. Specimen: The Golf Links, March 10, 1937. No. TR151.

ROSE FAMILY

Pyrus angustifolia Ait. (Narrow-leaved Crabapple)

Specimen: slopes of Tryon Mt., April 19, 1937. No. TR168.

Crataegus communis Beadle. (Hawthorn)

Common little shrub, holding its fruits much later than any other species. Specimen: In fruit, Oct. 14, 1937. No. TR91. *Fide*, Edw. Palmer.

Rubus argutus Link. (Blackberry). Open banks, Lynn. Specimen: May 16, 1937. No. TR190.

Rubus nigrobaccus Bailey. (Blackberry). Open banks, Pacolet Gorge. Specimen: May 6, 1937. No. TR179.

Rubus betulifolius Small. (Blackberry). Open banks, by the old Poinsett bridge, foot of Calahan Mt. Specimen: May 13, 1937. No. TR185.

The three species of *Rubus* were identified according to Small's new Manual. The treatment in Gray's Manual did not describe any of these specimens at all.

PEA FAMILY

Trifolium dubium Sibth. (Least Hop Clover)

In lawns; native of Europe. Specimen: April 30, 1937. No. TR176.

WOOD-SORREL FAMILY

Oxalis corniculata L. (Sorrel)

Oxalis repens Thumb.

Little weed, probably of foreign origin, in garden plots. Specimen: Oct. 27, 1936. No. TR102.

VINE FAMILY

Vitis Baileyana Munson

Trailing high in woods, "Valhalla". Specimen: In fruit, Oct. 2, 1936. No. TR35. *Fide*, Edw. Palmer.

ST. JOHN'S-WORT FAMILY

Ascyrum linifolium Spach. (St. Peter's-wort)

A low shrub, frequent in pine woods. According to Small's Manual this is a coastal plain plant. Specimen: Oct. 21, 1936. No. TR24. *Fide*, Edw. Palmer.

VIOLET FAMILY

Viola Lovelliana Brainerd.

Thinking the plants I found to be *V. triloba*, I took only a single specimen. On examination, however, it was found that the plant is pubescent only on the unfolding leaves and swiftly becomes glabrous. Every detail tallies with the description in Small's Manual and with the two drawings of *V. Lovelliana* in Brainerd's violet monograph (Vt. Agr. Expt. Sta. Bull. 224: pp. 33-35). A search of every species in Brainerd's monograph on violet hybrids (Vt. Agr. Expt. Sta. Bull. 239) fails to reveal anything that describes my plant, and it is to be remembered that Brainerd himself collected hybrids here assiduously in several years.

Viola Lovelliana is supposed to be confined to Louisiana, Arkansas and Oklahoma. That it has not been collected in intermediate states does not mean that it does not occur there, but only, perhaps, that collectors neglect violets as too difficult.

Specimen: Foot of Chimneytop Mt., April 28, 1937. No. TR169.

Viola Priceana Pollard (Confederate Violet)

In the *Flora of the Tryon Region*, following Brainerd, I considered this a mere albino, and a garden plant at that, of *V. papilionacea*. In the spring of 1937 however I discovered it in meadows where it was almost certainly wild. Further, Small has shown in his new Manual, p. 888, that the so-called Confederate Violet is a good species, while the albino form of *V. papilionacea* is an entirely different flower.

HEATH FAMILY

Gaultheria procumbens L. (Wintergreen)

Mrs. George Holmes showed me a small colony of this on the east slopes of Godshaw Hill. It has never been noted elsewhere in the county. Specimen: Leaves only, Nov. 27, 1936. No. TR148.

PRIMROSE FAMILY

Lysimachia Nummularia L.

Introduced from Europe; flowers in summer. This curious little plant is now naturalized well away from cultivation.

MILKWEED FAMILY

Vincetoxicum suberosum (L.) Britton (Angle-pod)

Gonolobus suberosus R. Brown

Cynanchum suberosum L.

Small in his Manual gives this only as a coastal plain plant. Twining on bushes. Specimen: "Valhalla", in fruit, Nov. 16, 1936. No. TR142.

MORNING GLORY FAMILY

Cuscuta indecora Choisy. (Dodder)

Cuscuta decora Engelm.

Collected July 20, 1933, near the Boy Scout Camp on Lake Lanier, but within the North Carolina boundary, by Budd Elmon Smith, who reports it in Elisha Mitchell Society Journal 50: 289-90, as new to the state. Smith reports it growing on legumes and asters.

Cuscuta compacta Jussieu. (Dodder)

Collected by Budd Elmon Smith (loc. cit.) from Polk Co. Specimen not seen.

PHLOX FAMILY

Phlox nivalis Loddiges. (Snowy Phlox)

This plant was collected by Miss Kimber in April, 1930, in the eastern part of the county. Specimen not seen by me, but identified by Dr. Wherry.

Phlox carolina L. (Carolina Phlox)

Considered by Dr. Edgar T. Wherry, our best authority, as distinct from *Phlox ovata* (which is also present), this is quite a common plant and remains remarkably long in flower. Specimen: Rocky Spur, Nov. 2, 1936. No. TR136.

POTATO FAMILY

Datura Stramonium L. (Stramonium, Jimson-weed)

A field weed, adventive from the American tropics. In fruit, March 18, 1935.

Physalis pubescens L. (Ground Cherry)

On dry sunny mountain fields. Specimen: Oct. 13 (in fruit), 1936. No. TR92.

MINT FAMILY

Pycnanthemum muticum Michx. (Mountain Mint)

Koellia mutica Britton.

Common in fields and along waysides. Specimen: Oct. 3, 1936. No. TR17.

COMPOSITES (ALL EXCEPT THE VERNAL IDENTIFIED
BY S. F. BLAKE)

Vernonia noveboracensis (L.) Willd. (Purple Ironweed)

In woods and fields. Specimen: Oct. 4, 1936. No. TR16.

Eupatorium incarnatum Walt.

A beautiful pink or pale purple flower. Frequent in cool glens. Specimen: Sept. 28, 1936. No. TR23.

Senecio aureus L. (Golden groundsel)

Common, open banks; the earliest of the senecios to bloom. Specimen: Fishtop, in fields, April 28, 1937. No. TR174.

Solidago Curtisi T. & G. (Golden-rod)

Common on sunny mountain slopes. The var. *pubescens* had already been collected here. Specimens: Oct. 14, 1936. Nos. TR68 and TR85.

Solidago petiolaris Ait. (Golden-rod)

On Melrose Mt. in deep woods. The var. *angustata* had already been collected here. Specimen: Oct. 14, 1936. No. TR80.

Solidago altissima L. (Golden-rod)

Roadsides, common. Specimen: Oct. 7, 1936. No. TR56.

Aster concolor L.

A beautiful and dainty little silver-leaved species found on sunny mountain slopes. Specimen: Oct. 14, 1936. No. TR76. This confirms my report in the *Flora of the Tryon Region* of its probable occurrence. In the *Flora* I also listed *Aster linariifolius* as having been reported, without specimens, by Miss Wright many years ago. This species also was confirmed by a specimen of that attractive little species which I took Oct. 14, 1936.

Aster Curtisi T. & G.

Roadsides, very common. Specimens: Oct. 7, Oct. 9, Oct. 17, 1936. Nos. TR40, TR51, TR65, TR88, TR91.

Aster pilosus Willd. var. *platyphyllus* (T. & G.) Blake.

Roadsides, common. Specimens: Sept. 28 and Oct. 7, 1936. Nos. TR24 and TR53.

Aster laevis L.

Frequent, along roadsides and in fields. Specimens: Oct. 7, 1936. No. TR43.

Baccharis halimifolia (Groundsel Tree)

This sea beach and salt marsh plant is certainly a surprising stray to find here. A large bush hangs over a roadside south of Clarke's Mt., in Greenville Co., S. C., in the extreme northeast square mile of the county. April 28, 1937. No. TR175.

Xanthium americanum Walt. (Cocklebur)

Xanthium pungens Wallr.

Xanthium pennsylvanicum Wallr.

Xanthium glabratum Britton

Xanthium canadense Mill.

A common field weed. Specimen: In fruit, Nov. 3, 1936.

Helenium tenuifolium Nutt. (Spanish Daisy)

A common and gay roadside weed. Specimen: Oct. 18, 1936. No. TR104.

Erechtites hieracifolia (L.) Raf. (Fireweed)

Roadsides and clearings, common. Specimens: Oct. 7 and Oct. 20, 1936. Nos. TR55 and TR120.

Cosmos bipinnatus Cav.

Native of tropical America, this well known garden flower is now so abundantly naturalized along roadsides that it must be admitted to the flora.

Hieracium scabrum Michx. (Hawkweed)

By roadsides and in fields. Specimens: Oct. 7, 1936. No. TR47.

Prenanthes serpentaria Pursh (Snakeroot)

In mountain woods. Specimen: Oct. 14, 1936. No. TR67.

Tragopogon pratensis L. (Goatsbeard)

Now a common wayside weed.

DELETIONS

The following species, reported in the *Flora of the Tryon Region* are there in error and must be excluded from the local flora:

Selaginella tortipila

The only voucher for this species in the local flora was one collected by Dr. Wherry, who has changed his identification of it to *S. rupestris*.

Tiarella macrophylla

According to Dr. Wherry this species is a chimera, the flowers on the type specimen, collected from the Tryon region, being those of typical *Tiarella cordifolia* while the leaves are those of *Heuchera villosa*.

Viola Stoneana

The specimens on which this report was based were misidentified by me and turn out to be several other sorts of violets.

Phlox divaricata

Reported in error.

Aster junceus

Specimens wrongly identified.

Antennaria occidentalis

Specimens wrongly identified.

CORRECTIONS

Introduction, p. 108, line 3; for "are", read: is

Introduction, p. 108, line 28; for "profit by" read: reward.

Species 4, line 1; for "Braken", read: Bracken.

Species 7, line 2; for "rare", read: frequent.

Species 62, line 2; after "and" add: waste.

Species 75, line 1; for "*Nothoholcus*," read: *Notholcus*.

Species 89, line 1; for "*berteroanus*," read: *Berteroanus*.

Species 106, line 3; for "specles", read: species.

Species 131, line 1, for "*halepense*", read: *halepensis*.

Species 203, delete lines 6 and 7.

Species 233, line 6; after 1927, add: (Wherry).

Species 255, line 2; for "aculis", read: acaulis.

Species 259, line 3; for "*Caiètes*", read: *Cleistes*.

Species 280, line 3; after Valhalla, add: (in fruit).

Species 285, line 1; for "K. Koch", read: C. Koch.

Species 307; this should be deleted, and the following substituted:

Celtis occidentalis L. (Hackberry)

Large tree, occasional in the mountain valleys. Our specimens differ in having garnet-red drupes and falcate leaves.

Species 316, line 10; for "kidnev," read: kidney.

Species 321, line 3; for "Britton", read: J. Britten.

Species 392, line 1; after *acuminata*, add: L.

Species 478, line 4; for "species", read: specimen.

Species 524, line 2; for "*Podalyriax*" read: *Podalyria*.

Species 637, line 1, after *Buckleyi*, insert: M. A. Curtis.

Species 678, line 5; for "May 5, 1911 (Tryon" read: Tryon, May 5, 1911 (Day).

Species 698, line 3; after *acutaefolium*, add: of Auths., not Michx.

Species 719, line 3; for "smal er", read: smaller.

Species 755, line 7; after 1897, add: (Towns.)

Species 753, line 4; after 1897, add: (Towns.)

Species 825, line 1, for "*clinopodia*", read: *Clinopodia*.

Species 831, line 4; for "*Ociganum*" read: *Origanum*.

Species 852, line 1; for "*Pentstemon*" read: *Penstemon*.

Species 862, line 4; for "Scherb." read: Schub.

Species 866, line 1; for "*austromantana*", read: *austromontana*.

Species 872, line 7; for "Valley", read: gorge.

Species 940, line 1; for "L." read: (L.) DC.

Species 959, line 3; for "in some places", read: as in some places.

Species 1008, line 2; for "*infrma*" read: *infirmus*.

Species 1010, line 2; for "*linifolius*", read: *linifolia*.

Species 1058, line 1; for "*Chicorium*", read: *Cichorium*.

Species 1059, line 2; for "*Taraxicum*", read: *Taraxacum*.

Species 1073; in the note appended, for "Tyron", read: Tryon.

Errata, species 614, for "line four," read: line 3.

Errata; delete completely the reference to species 720.

Errata, seventh line from the bottom of p. 156; for 848, read: 849.

List of New Names and Combinations (following Species 1073) species

380, for "Forma *Hugeri* Weatherby, read: Forma *laevicaulis* Harger.

Rhod. 31: 164. 1929.

THE MOSSES OF FRANKLIN COUNTY, NORTH CAROLINA

By LEWIS E. ANDERSON and EVELYN S. BEAVEN

An intensive study of the mosses of any small area is valuable. It not only adds to our knowledge of plant distribution but is helpful in establishing local habitat relationships and provides data that are often useful in making broader ecological generalizations. Franklin County was selected for the present study because it is situated on the borderline between the piedmont and coastal plain regions and thus offers excellent opportunities for studying the floristic transition between the two regions. This was the ultimate aim of the study.

Franklin County is located in the north-central part of North Carolina on the eastern edge of the Piedmont Plateau. The extreme eastern part lies within the coastal plain, while the western two-thirds is in the piedmont. In many places along the border of the two regions the sandy surface soil has been removed by erosion, thus exposing extensive areas of rock. These rock outcrops are characteristic of the transition region not only in Franklin County but in adjacent counties to the north and south as well. There are two main groups of soils in the county, the gray sandy surface soils, derived from upper coastal plain material and the red clay loam soils of the piedmont which are derived from crystalline rock. There is no sharp line of demarcation between these two types of soils. Isolated areas of gray sandy soil may appear in a section dominated by red clay while conversely small clay areas may be scattered within a more extensive sandy region.

Leo Lesquereux pointed out many years ago that mosses have their geological and lithological preferences far better marked than any other kind of plant. While there are many mosses which grow indifferently on a variety of substrata, there are some which are quite restricted as to habitat. It is thus possible to group the latter accordingly. In the present study nearly 600 collections of mosses were made within the county. As complete notes as possible were made regarding the habitat of each collection so that the full range of substrata for each species could be determined. The classification below follows that of Warming, which is based upon water requirements, i.e. into xerophytes, mesophytes, and hydrophytes. Further subdivision is necessary, however, in the case of mosses, to include the type of substratum.

XEROPHYTES: Plants which grow on substrata containing mostly very small amounts of water.

1. Plants growing on more or less exposed rocks.

Grimmia laevigata
Grimmia pilifera

Hedwigia ciliata
Ptychomitrium incurvum

2. Plants growing on the bark of living trees.

Clasmatodon parvulus
Drummondia prorrepens
Orthotrichum ohioense
Thelia hirtella

Cryphaea glomerata
Leptodon trichomitrium
Orthotrichum stellatum

3. Plants growing on soil, mostly old fields, roadsides, or open woods.

Pogonatum brachyphyllum
Bruchia Sullivanti
Dicranum condensatum
Ditrichum pusillum

Thelia Lescurii
Ditrichum pallidum
Polytrichum juniperinum

MESOPHYTES: Plants adapted to soil containing a moderate amount of water—essentially plants of shaded habitats.

1. Plants growing on moist shaded soil at the edge or banks of streams.

Bryhnia Novae-angliae
Eurhynchium hians

Atrichum undulatum
Mnium affine

HYDROPHYTES: Plants which are completely or partly submerged or which grow in very wet places.

1. Plants submerged or floating in water.

Aulacomnium heterostichum
Dicranum fuscescens
Tortella caespitosa
Plagiothecium micans
Leucobryum albidum
Polytrichum ohioense

Cirriphyllum Boscii
Campylium hispidulum
Eurhynchium serrulatum
Hypnum imponens
Hypnum curvifolium

2. Plants attached to rocks in water or wet by dripping or spraying water.

Fontinalis flaccida
Leptodictyum riparium var. *fluitans*

Fissidens Julianus

3. Plants growing in shaded woods, on soil at the base of trees, on humus or decaying wood.

Fissidens incurvus
Bryum bimum

Hygroamblystegium orthocladon
Hygroamblystegium irriguum

4. Plants growing in open bogs.

Aulacomnium palustre
Philonotis fontana
Hypnum arcuatum

Sphagnum imbricatum
Sphagnum subsecundum
Sphagnum imbricatum var. *affine*

5. Plants growing in more or less wooded swamps.

Climacium Kindbergii
Sphagnum (most species)
Brachythecium plumosum

Climacium americanum
Thuidium microphyllum
Campylopus chrysophyllum

Studies along the border of the coastal plain and piedmont failed to show any sharp distinction between the plants of the two regions. The only piedmont plants which cease abruptly at the line are those few species which grow exclusively on rocks, i.e. *Grimmia* spp., *Hedwigia ciliata*, and *Ptychomitrium incurvum*. That they do not occur in the coastal plain of Franklin County is due merely to the absence of rocks. In other counties of the coastal plain in which there are rocks these species are found in abundance. In the case of soil- and tree-growing species there is a general overlapping in the two regions. For instance, *Dicranum condensatum*, a typical plant of the sand hills of the coastal plain, was found growing throughout the county wherever there was any semblance of sandy soil of a xerophytic nature, irrespective of the two regions. Toward the western part of the county, however, it becomes less frequent and even rare due to the absence of suitable habitats. Other plants of the coastal plain exhibit this same type of distribution with a greater or lesser degree of overlapping. The species placed in this category are:

Sphagnum imbricatum
Pogonatum brachyphyllum
Funaria serrata

Atrichum crispum
Dicranum condensatum
Fontinalis flaccida

It should be noted that all of the above are plants which grow on soil, with the exception of *Fontinalis*. The epiphytes, as might be expected, are equally abundant in the two regions and none of the lower coastal plain species were found in Franklin County with the exception of *Entodon Drummondii* which was depauperate. Such plants as *Syrrophodon*

texanus, *Schlotheimia Sullivantii*, *Hypnum arcuatum* var. *americanum*, *Plagiothecium micans* var. *fulvum*, and *Dichelyma capillaceum* which are exceedingly abundant locally in the large swamps near the coast do not extend into Franklin County.

Of especial interest are the exposed rock outcrops referred to above which were studied in detail. Since the vegetation is essentially the same on all of these outcrops a description of one, known locally as "Flat Rock," will suffice. This outcrop is an extensive somewhat evenly eroded granitic exposure which slopes gradually from either side to the center through which a small stream flows. On each side of the stream the rock is dry and exposed to the sun during a large part of the day, thus offering an exceedingly xerophytic habitat. Areas of this nature support almost exclusively mats of *Hedwigia ciliata*, *Grimmia laevigata*, and *G. pilifera*, which, after a period of time retain a certain amount of soil. Where soil accumulation is sufficient the flowering plant, *Talinum teretifolium* Pursh, grows along with *Selaginella rupestris* (L.) Spreng. and several species of lichens. Soil is apparently built up in these otherwise bare areas in such a manner. Other species of flowering plants which eventually inhabit these mats are *Lonicera japonica* Thunb., *Bignonia capreolata* L., *Panicum* spp., *Solidago* sp., *Rhus copallina* L., and *Opuntia vulgaris* Mill. In the more moist situations along the stream *Anomodon attenuatus*, *Hypnum arcuatum*, and *Climacium americanum* flourish, often with mixtures of *Philonotis fontana* and *Atrichum undulatum*.

The check list of species collected in the county follows the arrangement of Grout in his Moss Flora of North America.

Order SPHAGNALES

Family SPHAGNACEAE

Sphagnum imbricatum Hornsch. On soil in boggy situations. Not common.

Sphagnum imbricatum var. *affine* (Ren. & Card.) Warnst. Bogs. Rare.

Sphagnum subsecundum Nees. Bogs. Common.

Order BRYALES

Family POLYTRICHACEAE

Atrichum angustatum (Brid.) Bryol. Eur. On soil in various situations, roadsides, old fields, open woods, etc. Common.

Atrichum crispum (James) Sull. Moist sandy soil, bank of streams. Rare.

Atrichum undulatum (Hedw.) Beauv. Moist soil, edge of streams, woods, fields, etc. Common.

Pogonatum brachyphyllum (Rich.) Beauv. Clayey banks. Not common.

Pogonatum pensilvanicum (Hedw.) Paris. Somewhat moist clay banks. Not common.

Polytrichum commune Hedw. Moist soil, swamps. Common.

Polytrichum juniperinum Hedw. In dry situations, often on rocks with a thin covering of soil. Uncommon.

Polytrichum ohioense Ren. & Card. Dry soil, woods, banks, roadsides, etc. Common.

Family FISSIDENTACEAE

Fissidens Bushii (Card. & Thér.) Card. & Thér. Clayey soils, banks, slopes, etc., in woods. Rare.

Fissidens Julianus (Mont.) Schimp. On rocks in running water. Not uncommon.

Fissidens taxifolius Hedw. Clayey soils, woods. Not common.

Fissidens viridulus (Web. & Mohr) Wahlenb. On rocks in streams. Common.

Fissidens viridulus var. *tamarindifolius* (Brid.) Grout. On stones in streams. Rare.

Family ARCHIDIACEAE

Archidium ohioense Schimp. Soil in old fields. Rare.

Family DITRICHACEAE

Pleuroidium subulatum (Hedw.) Lindb. Soil in old fields. Common.

Bruchia Sullivanti Aust. Old fields. Common.

Ceratodon purpureus (Hedw.) Brid. Dry soil on rocks, roadsides, and fields. Common.

Ditrichum pallidum (Hedw.) Hampe. On soil in various situations. Exceedingly common.

Ditrichum pusillum (Hedw.) E. G. Britton. Old fields and roadsides. Common.

Family DICRANACEAE

Dicranella heteromalla (Hedw.) Schimp. Soil, old fields, bank of streams, and roadsides. Common.

Dicranum condensatum Hedw. Dry sandy soil in open woods. Common.

Dicranum spurium Hedw. Shaded dry sandy soil. Common.

Dicranum scoparium Hedw. On soil, usually humus, in woods. Exceedingly common.

Dicranum fuscescens Turn. Decayed wood, woods. Common.

Dicranum flagellare Hedw. Decayed wood, shaded places. Not common.

Dicranum rugosum (Hoffm.) Brid. Humus in woods. Rare.

Family LEUCOBRYACEAE

Leucobryum albidum (Brid.) Lindb. Soil, decayed wood, and bases of trees. Common.

Leucobryum glaucum (Hedw.) Schimp. Soil in shaded places. Common.

Family POTTIACEAE

Weisia viridula Hedw. Dry soil. Exceedingly common.

Tortella caespitosa (Schwaegr.) Limpr. Soil and decaying wood, base of trees. Common.

Barbula unguiculata Hedw. On brick walls. Rare.

Acaulon triquetrum (Spruce) Muell. Soil in old field. Rare.

Tortula muralis Hedw. Brick walls. Rare.

Family GRIMMIACEAE

Grimmia laevigata (Brid.) Brid. Exposed rocks. Common.

Grimmia pilifera Beauv. Exposed rocks. Rather uncommon.

Hedwigia ciliata Hedw. Bare rocks. Common.

Ptychomitrium incurvum (Muhlenb.) Sull. Rock crevices. Common.

Family FUNARIACEAE

Physcomitrium turbinatum (Michx.) Brid. Moist soil, fields and pastures. Common.

Funaria flavicans Michx. Waste places and bare soil, especially burned-over areas, often mixed with the next. Not common.

Funaria hygrometrica Hedw. Same substrata as the last. Common.

Funaria serrata Brid. Moist sandy soils, banks and fields. Not common.

Family ORTHOTRICHACEAE

Orthotrichum ohioense Sull. & Lesq. Bark of trees. Common.

Orthotrichum stellatum Brid. Bark of trees. Not as common as the last.

Drummondia prorepens (Hedw.) Jennings. Bark of trees. Common.

Family BARTRAMIACEAE

Bartramia pomiformis Hedw. Soil in shaded places, occasionally on rocks. Common.

Philonotis fontana Brid. Soil and rocks in boggy places. Common.

Philonotis Muhlenbergii (Schwaegr.) Brid. Bogs and moist rocks. Rare.

Family AULACOMNIACEAE

Aulacomnium heterostichum (Hedw.) Bryol. Eur. On soil in woods, often at bases of trees. Common.

Aulacomnium palustre Schwaegr. Bogs and on rocks. Common.

Family BRYACEAE

Pohlia nutans (Hedw.) Lindb. On various substrata. Common.

Pohlia Wahlenbergii (Web. & Mohr) Andrews. Moist shaded soil. Not common.

Leptobryum pyriforme (Wils.) Schimp. Moist rocks. Rare.

Bryum argenteum Hedw. Soil, roadsides and waste places. Common.

Bryum bimum Brid. Wet rocks. Common.

Bryum caespitium Hedw. Soil, various places. Not common.

Bryum capillare Hedw. Moist situations, often bases of trees. Not common.

Rhodobryum roseum Schimp. Base of trees and stumps. Not common.

Family MNIACEAE

Mnium affine Schwaegr. Shaded soil in moist places. Common.

Mnium affine var. *ciliare* (Grev.) C. Muell. Moist soil, especially along streams. Common.

Mnium cuspidatum Hedw. Moist soil in woods. Common.

Mnium medium Bryol. Eur. Humus. Rare.

Mnium rostratum Schwaegr. Soil, shaded situations. Rare.

Family HYPNACEAE**Subfamily Climaciaceae**

Climacium americanum Brid. Moist shaded situations. Rather common.

Climacium Kindbergii (Ren. & Card.) Grout. Growing in or at the edge of water. Common.

Subfamily Beachythyetieae

Bryhnia Novae-Angliae (Sull. & Lesq.) Grout. Sandy soil, edge of streams. Rare.

Eurhynchium hians (Hedw.) J. & S. Moist shaded soil. Common.
Eurhynchium pulchellum (Hedw.) Jennings. Moist sandy soil.
Common.

Eurhynchium serrulatum (Hedw.) Kindb. Soil and humus in various situations. Exceedingly common.

Cirriphyllum Boscii (Schwaegr.) Grout. Shaded soil. Common.

Chamberlainia acuminata (Hedw.) Grout. Base of trees. Rare.

Brachythecium oxycladon (Brid.) J. & S. Soil, open woods and occasionally fields. Common.

Brachythecium plumosum Bryol. Eur. Soil, edge of streams. Not common.

Brachythecium salebrosum (Hoffm.) Bryol. Eur. Soil, banks, slopes, etc., in woods. Not common.

Subfamily Amblystegieae

Leptodictyum riparium (Hedw.) Warnst. Partially inundated rocks. Not uncommon.

Leptodictyum riparium var. *fluitans* Lesq. & James. In running water. Rare.

Leptodictyum riparium var. *longifolium* Schultz. Stagnant water. Rare.

Amblystegium varium (Hedw.) Lindb. Various substrata. Common.

Hygroamblystegium irriguum (Wils.) Loeske. In wet soil and in water. Common.

Hygroamblystegium orthocladon (Beauv.) Grout. In streams. Common.

Campylium chrysophyllum (Brid.) Bryhn. On soil and rocks with a thin layer of soil. Common.

Campylium chrysophyllum var. *brevifolium* (Ren. & Card.) Grout. On soil, edge of streams. Common.

Campylium hispidulum (Brid.) Mitt. Decaying wood, stumps, etc. Not uncommon.

Drepanocladus aduncus (Hedw.) Warnst. var. *Kneiffii* (Bryol. Eur.) Warnst. In bogs and swampy situations. Not common.

Subfamily Hypneae

Hypnum arcuatum Lindb. In boggy situations. Common.

Hypnum curvifolium Hedw. Decaying wood and soil in shaded places. Not common.

Hypnum imponens Hedw. Soil and decaying wood. Uncommon.

Hypnum molluscum Hedw. Soil, bases of trees. Rare.

Heterophyllum Haldanianum (Grev.) Kindb. Moist decaying wood.
Rare.

Sematophyllum adnatum (Michx.) E. G. Britton. On bark of living trees. Common.

Sematophyllum carolinianum (C. Muell.) E. G. Britton. Moist rocks.
Rare.

Homomallium adnatum (Hedw.) Broth. Bark of trees, decaying wood, and rocks. Not common.

Pylaisia intricata (Hedw.) Bryol. Eur. Bark of living trees. Not common.

Pylaisia Selwynii Kindb. On trees. Uncommon.

Platygyrium repens (Brid.) Bryol. Eur. Decayed wood. Common.

Subfamily Plagiothecieae

Plagiothecium denticulatum (Hedw.) Bryol. Eur. On soil and humus in shaded places. Not common.

Plagiothecium micans (Sw.) Paris. Decaying wood. Common.

Subfamily Entodontae

Entodon cladorrhizans (Hedw.) C. Muell. Moist rocks. Rare.

Entodon Drummondii (Bryol. Eur.) J. & S. Bark of tree. Rare.

Entodon seductrix (Hedw.) C. Muell. Soil and rocks. Common.

Family LESKEACEAE

Thuidium delicatulum (Hedw.) Mitt. Various substrata. Common.

Thuidium microphyllum (Hedw.) Best. Soil and decaying wood in woods and swampy situations. Common.

Thuidium virginianum (Brid.) Lindb. Soil in rather dry situations.
Not common.

Leskea gracilescens Hedw. Bark of trees and decaying logs with soil.
Not common.

Leskea obscura Hedw. Bark of trees. Common.

Thelia asprella Sull. Base of trees. Common.

Thelia hirtella (Hedw.) Sull. Tree trunks. Rare.

Thelia Lescurii Sull. Sandy soil and rocks with a thin layer of soil.
Not uncommon.

Anomodon attenuatus (Hedw.) Hueben. Soil in bottom land and at bases of trees and rocks. Common.

Anomodon rostratus (Hedw.) Schimp. Bases of trees and rocks.
Common.

Family LEUCODONTACEAE

Leucodon julaceus (Hedw.) Sull. Bark of trees. Common.

Leptodon trichomitrium (Hedw.) Mohr. Bark of trees. Not common.

Family CRYPHAEACEAE

Cryphaea glomerata Schimp. Bark of trees. Common.

Cryphaea nervosa (Hook. & Wils.) Bryol. Eur. Bark of trees, often mixed with the last. Rare.

Family FABRONIACEAE

Clasmatodon parvulus (Hampe) Sull. Bark of trees and decaying wood. Common.

Schwetschkeopsis denticulata (Sull.) Broth. Bark, bases of trees. Rare.

Family FONTINALACEAE

Fontinalis flaccida Ren. & Card. In water. Not common.

The writers are indebted to Professors H. L. Blomquist and H. J. Oosting for helpful suggestions and criticisms during the course of the work.

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THE MORPHOLOGY OF THE PERITHECIUM OF SORDARIA FIMICOLA (ROB.) CES. AND DE NOT.

By DON RITCHIE

PLATES 28 AND 29

The first important work on the Sordariaceae was done by Dangeard (8) in 1907. He decided for *Sordaria fimicola* that there was no antheridium and no fertilization, that the cells of the ascogonium were multinucleate, and that the asci were produced from the ascogonium by crozier formation. He made no further observations on the nuclear behaviour of the fungus. *Sordaria macrospora* was described as essentially similar except that it has a straight, intercalary archicarp instead of a coiled, terminal one. Only in *Sporormia intermedia* he did describe binucleate cells in the ascogonium and even went so far as to say "One would be tempted . . . to think of a fusion."

Satina (17) has also contributed important developmental studies on the Sordariaceae. In the several members of this family considered, she describes the same multinucleate, coiled ascogonium, without an antheridium, and the ascogenous hyphae coming from the ascogonium. Paired nuclei were seen, but not in the cells which formed asci. Large nuclei and small nuclei were observed, but fusion was denied, the large nuclei being explained as "fully grown, ready for dividing."

MATERIALS AND METHODS

The material was collected on horse and cow dung and was got into pure culture by cracking the perithecia and placing the living masses of asci on agar plates. When the hyphae outgrew the bacteria, the clean ends were cut off and transferred to other plates. For this purpose various agars were used. Blakeslee's #230 proved to be the most satisfactory for the production of perithecia, as it becomes black with fruit about a week after inoculation. Prune decoction agar gives a heavy vegetative growth but fruiting is poor. On maltose-peptone, potato-dextrose, and corn-meal-yeast agar, *Sordaria fimicola* grows well, but not as luxuriously as on #230. Dung agars made from decoctions of cow, horse, or sheep dung, did not give any better growth than that

which was obtained on plain, non-nutrient agar. Cultures were also made by letting the perithecia shoot spores onto agar plates. Germination was attempted on all the agars on which the fungus was grown, but the spores did not germinate on any except plain, non-nutrient agar. Some of the spores were placed for several hours in the sunlight, but without an increase either in the rapidity or percentage of germination. Various temperatures were used, as suggested by Dodge (9), but without success. However, on plain, non-nutrient agar, the spores will germinate readily either in the light or darkness at room temperatures (15-25 degrees C.).

Perithecial development was studied in living material until the perithecia became too opaque to observe. There is a period, as noted by Gilkinet (13), during which living material is worthless for study, as the thickening wall of the perithecium cannot be opened without destroying the contents. During this period (and this is just when the most interesting developments are taking place) sections must be used. For this purpose, the fungus was killed in various fixers. Carnoy's fluid caused plasmolysis. Chromic formalin and hot corrosive sublimate in 95% alcohol gave indifferent results. The Chicago formula (6) used instead of Flemming's weaker solution because of the lesser percentage of osmic acid, proved most satisfactory. Because of the air entrapped in the interstices of the aerial hyphae, the material to be fixed had to be kept under the aspirator for six to twelve hours. Zirkle's (18) N-butyl series of alcohols was used for dehydration and infiltration. Sections were cut at 5 μ , and stained with Gram's gentian violet and iodine and with Heidenhain's iron alum haematoxylin, counterstained in Orange G, light green or fast green. The haematoxylin alone gave the clearest nuclear figures, while this stain followed by Orange G was most useful in bringing out the arrangement of the cells in the base of the perithecium.

THE MYCELIUM

The spores of *Sordaria fimicola* germinate by means of an apical germ pore, as shown by Gilkinet (13). A vesicle is given out, round and full of cytoplasm, from which proceed one to several hyphae (Fig. 1). These hyphae become septate, with perforations in the septa. The mature mycelium is composed of hyphae of various diameters, ranging from 3 μ to 12 μ in diameter. On media rich in nourishment, many aerial hyphae are produced, but on less nutrient, the mycelium keeps to the surface or burrows into the substratum.

Perithecia with eight-spored asci are formed in single-spore cultures, and thus the fungus is homothallic.

THE ARCHICARP

At the time of the formation of the perithecial fundaments, as shown by previous workers on the *Sordarias* (Gilkinet, 13; Dangeard, 8; Satina, 17) certain of the ends of otherwise undifferentiated hyphae become coiled (Figs. 2 and 3). Septations appear (Fig. 4), but not with any apparent regularity such as Andrus (2) found for *Ceratostomella*. This coil, or archicarp, twists about itself for several turns. One or more hyphae may contribute to the formation of the archicarp (Figs. 2 and 3), which soon becomes invested with smaller, numerous vegetative hyphae (Fig. 5). Neither Dangeard (8) nor Satina (17) considered any of these as antheridia, and there is indeed no indication that any of them function in a sexual way. Usually there is no structure that can be interpreted as a trichogyne, though there appear occasionally unaccountable out-growths from the archicarp (Fig. 4). Neither are there any conidia such as Satina (17) shows for *Podospora curvula*, nor any spermatia such as Ames (1) describes for *Pleuraea anserina*.

PERITHECIAL DEVELOPMENT

After the investment of the archicarp by the vegetative hyphae, the entire mass grows quickly, soon becoming too opaque to study. At this time the covering hyphae are so strong, and the contents so delicate, that crushing the perithecia destroys the inner cells. In sectioned material, however, it can be seen that the archicarp has grown into a homogeneous tangle of filaments which are already becoming pseudoparenchymatous in nature (Fig. 6). Even in the early stages, the outer layer of cells is heavier-walled than the internal portion. When the perithecial covering is still only one layer thick, there becomes differentiated in the center of the previously homogeneous tissue, a more deeply staining hypha (Fig. 7) which seems to be what Fuisting (12) described for *Stictosphaeria*, *Diatrype*, etc., as the Woronin hypha. Fisch (11), who considered Fuisting's paper "downright foggy," said, "It can not be doubted that the Woronin hypha arises as a new formation in the matrix, though its rise cannot be followed." He states that the Woronin hypha stains heavily, has very delicate walls, and grows to several times its original size. For *Xylaria*, however, Fisch (l.c., p. 879) rejects Brefeld's assertion that the asci arise from the Woronin hypha.

Similar organisms (in which the archicarp merely produces vegetative

growth) have been noted by several investigators. Nichols in 1896 (16) on *Hypocopa* found this to be true. Blackman and Wolsford (3) in 1912 found sexually non-functional archicarps in *Polystigma rubrum*, Jones in 1926 (15) noted a like situation in *Ophiobolus graminis*, as did Cookson in 1928 (17) on *Melanospora zamiae*.

As development proceeds, the thin-walled cells forming the center of the perithecium begin to disintegrate, and the cells of the outer layers become stouter, the outermost being darkly pigmented. In living material at this stage, many elliptical or roundish, thin-walled, apparently empty chains of cells can be seen, as Gilkinet (13) shows. These are probably nourishing cells, and are so delicate that they never keep their shape in fixed material. In sectioned perithecia these nourishing cells seem to be disintegrating progressively from the center to the outside (Figs. 10 and 12).

Now the deeply staining hypha (or hyphae) in the center of the perithecium appears to be broken up into numerous cells. Though never more than one hypha was seen at one time in a perithecium, it would be hard to say whether one or more hyphae are involved in this procedure because the resulting cells are so irregularly disposed (Figs. 9 and 13). The cells round off and separate from one another in a fashion similar to that found in *Xylaria* by Brown (4) and in *Ceratostomella* by Andrus (2). These irregularly arranged, deeply staining cells make up the ascogenous hyphae. Some of the cells are uninucleate, many are binucleate, and a few have three nuclei. These nuclei, like those Jones (15) found in *Ophiobolus*, show no internal detail.

DEVELOPMENT OF THE ASCOGENOUS HYPHAE

In *Sordaria fimicola* the paired nuclei fuse in the primary ascus cell (Figs. 13 and 14). The nuclei before fusion are considerably smaller than the fusion nucleus. Following fusion in the primary ascus cell, the ascus grows upward without forming the typical crozier, though occasionally regular ones are found (Figs. 11 and 15). This situation in *Sordaria fimicola* is substantiated by Faull (10) and is paralleled in *Ophiobolus graminis* as described by Jones (15), who says that typical croziers are sometimes formed and sometimes not. The same is true for *Cordyceps agariciformia*, according to Jenkins (14). Cookson (7) on *Melanospora zamiae*, Brown (4) on *Xylaria*, and Cayley (5) on *Nectria galligena* all describe the formation of asci from ascogenous cells without croziers.

Thus *Sordaria fimicola* is added to a growing list of species which

form asci from purely vegetative hyphae, a list which began with the study of *Xylaria* by Fisch (11) in 1882, and which now includes members of the Pseudosphaeriaceae, Hypocreales, Sordariaceae, Ceratostomaceae, Gnomoniaceae, Xylariaceae, and Pezizaceae.

STRUCTURE OF THE PERITHECIUM

After the fusion of the nuclei in the ascogenous cells, the asci increase greatly in size and remain uninucleate until they have reached their full growth. Meanwhile, the layers of the perithecial wall have differentiated somewhat. The outermost layer, one cell thick, is heavily pigmented but fairly thin-walled. The layer next to the outside is several cells thick, and consists of extremely thick-walled cells. Inside that, a few layers of thin-walled cells form the lining of the perithecium. At the top of the perithecium, opposite the origin of the asci, the perithecial wall elongates, and the cells of the inner wall grow usually upward and outward, forming the periphyses, and making an opening, the ostiole, through what has by now become the neck.

CYTOLOGY OF THE ASCUS

The young ascus is distinguished by a mass of darkly staining material in the tip, and by the large fusion nucleus (Fig. 18). The dark material is seen in even the smallest asci in which fusion has taken place, and remains until the formation of the ring in the tip of the mature ascus. The fusion nucleus is relatively large, having a diameter half as great as the diameter of the ascus. It has a conspicuous nucleolus surrounded by a clear area. Outside the membrane are aggregated some dark masses of material, not distinct, but somewhat granular in nature, which are seen only at this time. Faull (10) also observed this. The cytoplasm of the ascus is denser at this time than at any later time, but is characteristically vacuolate at the lower end.

The first division in the ascus occurs with the spindle more or less parallel to the long axis of the ascus (Fig. 19). The chromosomes are too small and indistinct to be counted. The nucleolus remains at least through the metaphase as a large, distinct, dark body alongside the chromosomes.

Since there are comparatively few asci found between the time of the first mitosis and the delimitation of the spores, the remaining stages in ascus formation must pass rapidly. The nuclei resulting from the first division are smaller than the fusion nucleus, but still contain a prominent nucleolus, located in the nucleus in such a way that each nucleus

seems to have a beak pointing away from the other nucleus (Fig. 21). The second division follows with its spindle also practically parallel to the long axis of the ascus (Fig. 22). The division figure is essentially similar to the first, though smaller and with less conspicuous nucleoli. In the four-nucleate stage, the nuclei are still smaller, but they retain the characteristic appearance, with a distinct nucleolus surrounded by the clear area. A chromatin network was seen in resting nuclei only in the binucleate stage. At the end of the second division, the ascus has completed its cytoplasmic development. The inner wall of the ascus is distinct, and the ring in the tip of the outer wall is complete (Fig. 23).

The spindles of the third mitosis lie somewhat transverse to the long axis of the ascus (Fig. 24). They are long and quite slender, with still smaller nucleoli, and when complete, extend from side to side of the ascus. When the nuclei are entirely separated, they send out beaks at the point of which are located the centrosomes. From these centrosomes radiate the astral rays that delimit the spores. There is still a nucleolus present on the nucleus (Fig. 26). The spore membrane remains quite delicate until the nucleus has retreated from the wall and undergone a last mitosis (Fig. 27) which leaves the finished spore binucleate (Fig. 28).

SUMMARY

1. *Sordaria fimicola* (Rob.) Ces. and De Not. is a homothallic member of the coprophilous Sphaeriales, as indicated by cultures descended from single spores.

2. The perithecial initial is a coiled, septate, terminal hypha, which becomes invested with twining vegetative hyphae. There is no antheridium.

3. The original archicarp forms vegetative hyphae only. There arises *de novo*, a darkly staining hypha which appears to give rise to a tangle of uninucleate and binucleate cells that round off, separate, and increase in size.

4. The paired nuclei in these ascogenous cells fuse, and the cells proceed to ascus formation, sometimes with but usually without typical crozier formation.

5. The observations on the formation of the perithecial wall and neck agree with those of Woronin, Gilkinet, and Dangeard.

6. Observations on the behavior of the nuclei in the ascus, and on the formation of the ascospores agree with those of Faull.

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LITERATURE CITED

1. AMES, L. M. An hermaphroditic self-sterile but cross-fertile condition in *Pleurage anserinus*. Bull. Torr. Bot. Club 59: 341-345. 1932.
2. ANDRUS, C. F. Cell relations in the perithecium of *Ceratostomella multiangulata*. Mycologia 26: 133-153. 1936.
3. BLACKMAN, V. H., AND WELSFORD, E. J. The development of the perithecium of *Polystigma rubrum*, DC. Ann. Bot. 26: 761-767. 1912.
4. BROWN, H. B. Studies in the development of *Xylaria*. Ann. Mycol. 11: 1-13. 1913.
5. CAYLEY, DOROTHY M. Some observations on the life-history of *Nectria galligena*, Pers. Ann. Bot. 35: 79-92. 1921.
6. CHAMBERLAIN, C. J. Methods in Plant Histology. 5th ed. University of Chicago Press. Chicago, 1932.
7. COOKSON, ISABEL. The structure and development of the perithecium in *Melanospora zamiae*, Corda. Ann. Bot. 42: 255-269. 1928.
8. DANGEARD, P. - A. Recherches sur le développement du Périthèce chez les Ascomycètes. Le Botaniste 10: 1-385. 1907.
9. DODGE, B. O. Methods of culture and the morphology of the archicarp in certain species of the Ascobolaceae. Bull. Torr. Bot. Club 39: 139-197. 1912.
10. FAULL, J. H. Development of the ascus and spore formation in the Ascomycetes. Proc. Boston Soc. Nat. Hist. 22: 77-114. 1905.
11. FISCH, C. Beiträge zur Entwicklungschichte einiger Ascomyceten. Bot. Zeit. 40: 851-870; 875-897; 899-906. 1882.
12. FUISTING, W. Zur Entwicklungschichte der Pyrenomyceten. Bot. Zeit. 25: 177-181; 185-189; 193-198. 1867.
13. GILKINET, ALFRED. Recherches morphologiques sur les Pyrénomycètes. I. Sordariées. Bull. Acad. Roy. de Belge, 2nd ser. 37, part 1: 426-448. 1874.
14. JENKINS, W. A. The development of *Cordyceps agariciformis*. Mycologia 26: 220-243. 1934.
15. JONES, S. G. The development of the perithecium of *Ophiobolus graminis*, Sacc. Ann. Bot. 40: 607-629. 1926.
16. NICHOLS, MARY A. The morphology and development of certain pyrenomycetous fungi. Bot. Gaz. 22: 301-328. 1896.
17. SATINA, S. Studies in the development of certain species of the Sordariaceae. Bull. Soc. Nat. Moscow 30: 106-142. 1916.

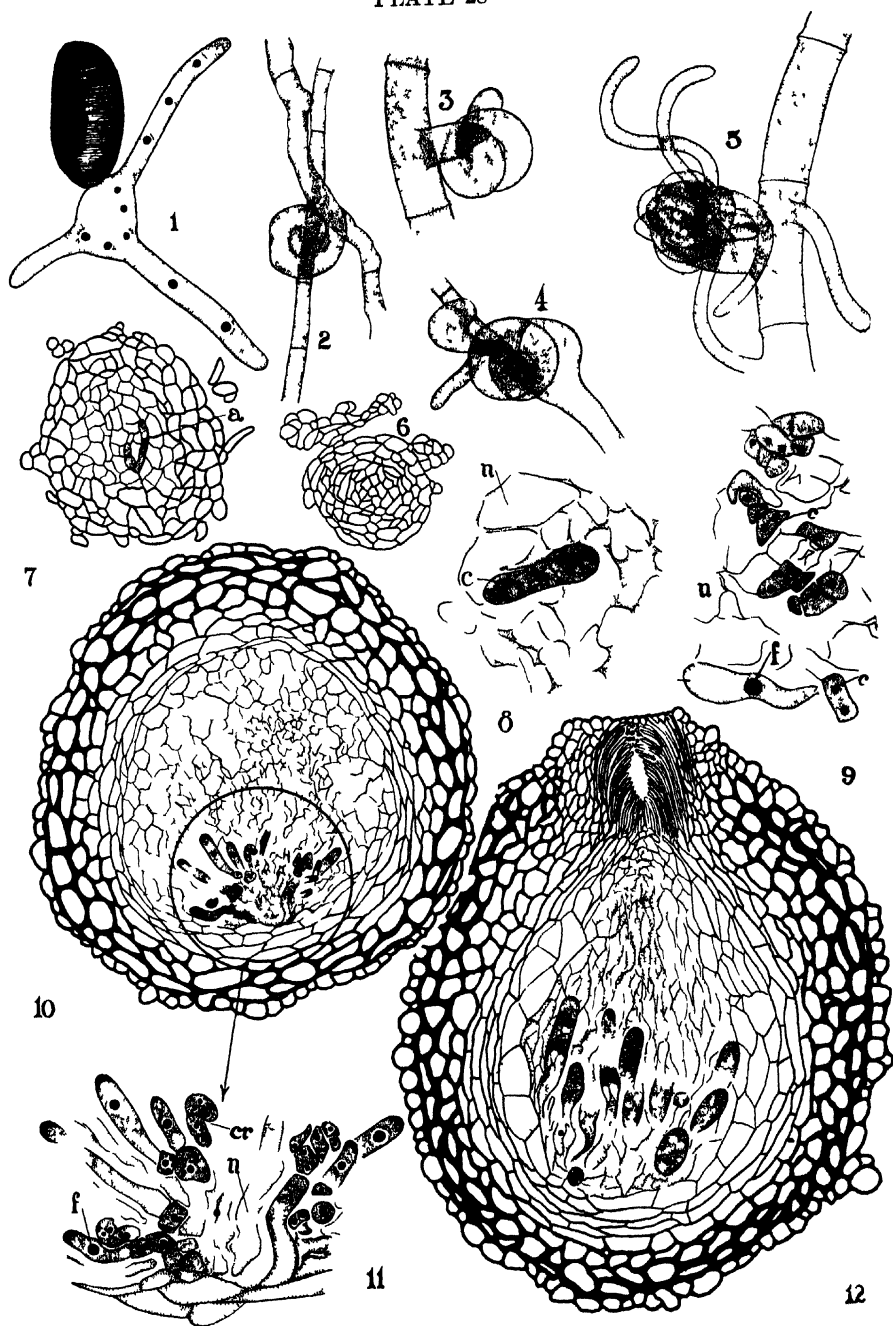
18. ZIRKLE, C. The use of N-butyl alcohol in dehydrating woody tissue for paraffin embedding. *Science* 71: 103-104. 1930.

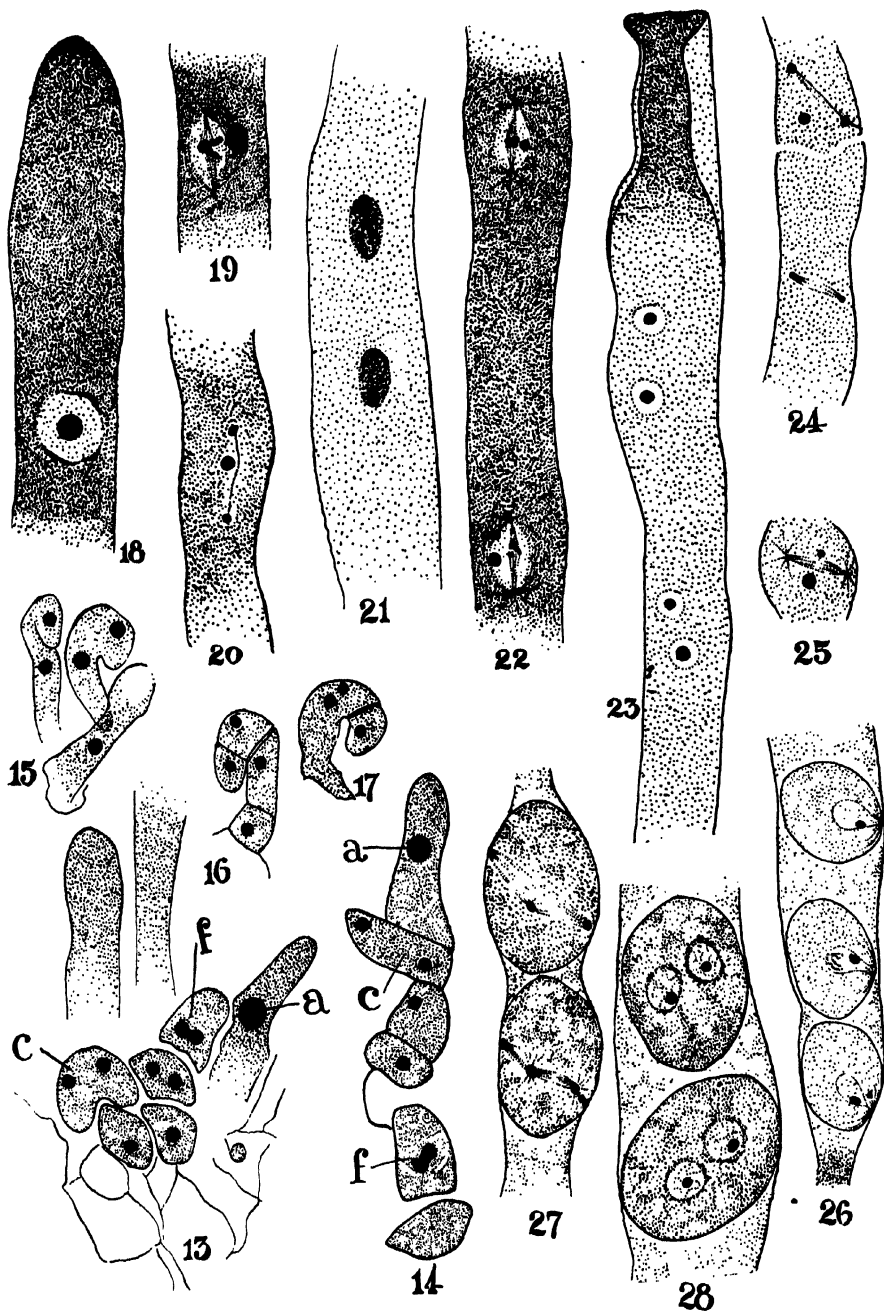
EXPLANATION OF PLATES 28 AND 29

1. Germinating spore, $\times 1135$.
2. Archicarp originating from several hyphae, still non-septate, $\times 1135$.
3. Archicarp originating as a side branch of a hypha, $\times 1135$.
4. Archicarp with septations, $\times 1135$.
5. Archicarp surrounded with vegetative hyphae, $\times 1125$.
6. Section through a young perithecial fundament showing homogeneous nature of the tissue, $\times 360$.
7. Section through a little older perithecial fundament showing beginning of differentiation, with darker staining hypha in the center, $\times 360$.
8. Portion from section of a still older perithecium. The dark cells form an ascogenous hypha, while the irregular cells are the disintegrating nourishing tissue, $\times 1135$.
9. Portion of section of perithecium only a little younger than that in Fig. 10. Binucleate cells (c), nourishing cells (n), and a fusion nucleus (f) are to be seen, $\times 1135$.
10. Entire section through a young perithecium, showing disintegration of the central cells, thickening of the walls, and rise of the ascogenous hyphae from the base of the perithecium, $\times 360$.
11. Enlarged portion of Fig. 10, showing binucleate cells, nourishing cells (n), fusion nuclei (f), and a crozier (cr). The origin of the asci from the vegetative cells can be traced, $\times 745$.
12. Section through an almost mature perithecium. The uninucleate asci are nearly full grown. The walls are complete and the neck is rapidly being formed, $\times 360$.
13. Ascogenous hyphae from a young perithecium. The asci are just being formed. Here are shown the binucleate cells (c), nuclei fusing (f), and the fusion nucleus (a) of the young ascus.
14. Another example of the same stage.
15. Early steps in crozier formation.
16. Later stages in crozier formation. One nucleus of the primary ascus cell has presumably been cut away.
17. An irregular crozier, in which one wall has not been laid down.
18. Tip of young ascus with large fusion nucleus.
19. Metaphase of first mitosis.
20. Telophase of first mitosis.
21. Portion of a binucleate ascus, showing the nucleoli at opposite sides of the nuclei.
22. Metaphase of the second mitosis.
23. A quadrinucleate ascus. The double wall of the ascus is visible at the distal end. The thickening in the tip is the ring in the outer wall of the ascus.
24. Telophase of the last mitosis. Only two of the mitoses are visible.
25. Another example of the same stage, with astral rays apparent.
26. Spores being delimited on the astral rays from the centrosomes of the beaked nuclei.

27. The last mitosis occurring in the completely delimited spore.
28. Spores complete except for the laying down of the heavy outer wall.

Figs. 1-17 were stained in Haidenhain's iron alum haematoxylin and Orange G. Figs. 18-28 were stained in haematoxylin alone. All magnifications on Plate 29 are $\times 2270$, others as indicated.





OBSERVATIONS ON THE REPRODUCTION OF ANCYLISTES*

By HELEN BERDAN

Two species of *Ancylistes*, the type genus of the order Ancylistales, were found parasitizing species of *Closterium* from a pool in Sparrow's cow pasture, Chapel Hill, N. C., during the summer of 1937. One of these species, *A. Closterii*, was originally reported and described by Pfitzer in 1872. It has since been found by Dangeard, Sorokine, Wildeman, Constantineanu, Petersen, Schultz-Danzig, and Valkanov in Europe and Asia. The presence of this fungus in America has been noted in unpublished records of Harvey (Mississippi, 1924) and Couch (North Carolina, 1925, 1932, 1934). The second species, *A. Pfeifferi*, was discovered in Brazil and added by Beck in 1897 to the order Ancylistales established by Schröter in that same year. This species was first seen by the writer in *Closterium* species taken from a bog near London, Canada, in 1935. The presence of nine to ten blunt projections from the wall of the cell enclosing the zygote or resting-spore readily distinguishes it from *A. Closterii* in which this cell is smooth-walled.

As a result of a critical study of these two species from the Chapel Hill material, undertaken at the University of North Carolina this summer, a paper, now in press, deals with the revision of the genus *Ancylistes*. The primary basis of the revision lies in the discovery of asexual reproduction by conidia. Until now, *Ancylistes* has been separated from the other members of the order by its characteristic method of infecting by narrow tubular, external hyphae rather than by zoospores. It has now been found that these hyphae function as infection tubes only when they are entirely submerged in water. Those which emerge from water into air function as conidiophores. The conidia resemble those of *Conidiobolus* sp. (Couch) both in structure and forcible mode of discharge. Germination may produce secondary conidia or germ tubes. The germ tubes may function directly as infection tubes or as conidiophores as described by Martin for *Conidio-*

*A fuller treatment with illustrations and bibliography will appear in *Mycologia* in 1938.

bolus villosus. Secondary conidia may form tertiary conidia, be forcibly ejected, or germinate *in situ* by tube.

Each external hypha, which is potentially either an infection tube or a conidiophore, develops from a single internal mycelial segment. The whole protoplast of the intramatrical segment passes out through the wall of the host cell into the external hypha and flows into its growing tip by progressive stages. According to conditions, this protoplast eventually becomes concentrated in a swollen end cell of the infection hypha attached to a new host as described by Pfitzer and others, or in the conidium as observed by the writer. The content of the conidium, upon germination by tube, collects in a similar swollen infection cell. From this attached end cell of the infection hypha or the germ tube penetration of the new *Closterium* cell occurs by a fine, conical papilla. Once inside the host, the papilla elongates into a narrow tube. Through this the protoplast of the infection cell flows rapidly to form an ovoid ball in the cytoplasm of the host. Elongation into a continuous intramatrical hypha ensues. Later the hypha septates, and at maturity all the cells become either external hyphae or gametangia.

The thalli of both *A. Closterii* and *A. Pfeifferi* appear to be homothallic, conjugation being either lateral or scalariform. This disagrees with previous accounts which claim heterothallism with an appreciable difference in the diameter of the male and female filaments. Sexual reproduction appears to be mainly zygomycetous in character. The mature zygote or resting-spore, however, is seen to be retracted from the wall of the cell in which it is formed. This conforms to the view of Brefeld that sexual reproduction in *Conidiobolus* is intermediate between the true zygomycetous and oomycetous types.

The discovery of forcibly ejected conidia and lateral conjugation seems to warrant a revision of the genus *Ancylistes* and its removal from the Ancylistales to the Entomophthorales in a position near *Completothoria complens* Lohde.

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